Clinical Study Protocol
Sponsor:
GlaxoSmithKline Biologicals
Rue de l'Institut, 89
1330 Rixensart, Belgium

Primary Study vaccine and number	• GlaxoSmithKline (GSK) Biologicals' respiratory syncytial virus (RSV) vaccine based on the RSV viral proteins F, N and M2-1 encoded by chimpanzee-derived adenovector (ChAd155-RSV) (GSK3389245A)
Other Study vaccine	• Placebo (Formulation buffer S9b)
eTrack study number and Abbreviated Title	204838 (RSV PED-002)
Investigational New Drug (IND) number	16999
EudraCT number	2016-000117-76
Date of protocol	Final Version 1: 26 April 2016
Date of administrative change	Administrative change 1 Final: 06 July 2016 Administrative change 2 Final: 13 September 2016
Date of protocol amendment	Amendment 1 Final: 16 January 2017 Amendment 2 Final: 08 June 2017 Amendment 3 Final: 12 September 2017 Amendment 4 Final: 10 December 2017
Title	A study to evaluate safety, reactogenicity and immunogenicity of GSK Biologicals' RSV investigational vaccine based on viral proteins encoded by chimpanzee-derived adenovector (ChAd155-RSV) (GSK3389245A) in RSV-seropositive infants.
Detailed Title	A Phase 1/2, randomized, observer-blind, controlled, multi-center, dose-escalation study to evaluate safety, reactogenicity and immunogenicity of GSK Biologicals' respiratory syncytial virus (RSV) investigational vaccine based on the RSV viral proteins F, N and M2-1 encoded by chimpanzee-derived adenovector (ChAd155-RSV) (GSK3389245A), when administered intramuscularly according to a 0, 1-month schedule to RSV-seropositive infants aged 12 to 23 months.

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Co-ordinating authors	• PPD (Expert Scientific Writer) • PPD (Scientific Writer, Keyrus Biopharma contractor for GSK Biologicals) • PPD, Scientific Writer
Contributing authors (Amended 10 December 2017)	• PPD (Clinical Research and Development Lead [CRDL]) • PPD (Clinical and Epidemiology Research & Development Project Lead) • PPD (Lead Statistician) • PPD (Lead Statistician) • PPD (Lead Statistician) • PPD ***(Project Statistician)*** • PPD (Project Statistician) • PPD (Project Statistician) • PPD (Study Delivery Lead) • PPD (Study Delivery Lead) • PPD (Study Delivery Manager, Synteract HCR contractor for GSK Biologicals) • PPD (Vaccine Supply Coordinator, CVO-Europe contractor for GSK Biologicals) • PPD (Vaccine Supply Coordinator)

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Contributing authors (continued) (Amended 10 December 2017)	• PPD ***, (Clinical Trial Supply Manager)*** • PPD (Clinical Trial Supply Manager, Aixial contractor for GSK Biologicals) • PPD (Clinical Read-out Team Leader) • PPD (Clinical Laboratory Sciences [CLS] Study Manager, Business & Decision Life Sciences contractor for GSK Biologicals) • PPD ***(Central Safety Physician)*** • PPD ***(Safety Scientist)*** • PPD (Central Safety Physician) • PPD (Project Data Manager) • PPD (Portfolio Data Manager) • PPD (Oversight Data Manager) • PPD (Oversight Data Manager, Keyrus Biopharma contractor for GSK Biologicals) • PPD (Global Regulatory Representative) • PPD (Global Regulatory Representative) • PPD (Global Regulatory Representative) • PPD (Global Patent Representative)

GSK Biologicals' Protocol DS v 14.1.1

Protocol Amendment 4 Sponsor Signatory Approval

eTrack study number and Abbreviated Title	204838 (RSV PED-002)
IND number	16999
EudraCT number	2016-000117-76
Date of protocol amendment	Amendment 4 Final: 10 December 2017
Detailed Title	A Phase 1/2, randomized, observer-blind, controlled, multi-center, dose-escalation study to evaluate safety, reactogenicity and immunogenicity of GSK Biologicals' respiratory syncytial virus (RSV) investigational vaccine based on the RSV viral proteins F, N and M2-1 encoded by chimpanzee-derived adenovector (ChAd155-RSV) (GSK3389245A), when administered intramuscularly according to a 0, 1-month schedule to RSV-seropositive infants aged 12 to 23 months.
Sponsor signatory	Amanda Leach, Clinical and Epidemiology Research & Development Project Lead
Signature	_____
Date	_____

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Protocol Amendment 4 Rationale

Amendment number:	Amendment 4

Rationale/background for changes:

- An Independent Data Monitoring Committee (IDMC) is reviewing all accumulating unblinded safety and reactogenicity data on a monthly basis for this study to ensure that there is a timely identification of any safety signal. As safety data accumulates, it may be that there is sufficient evidence of safety of the current dose level to allow progression to the next dose level. For instance, taking the *a priori* safety concern of thrombocytopenia, this amendment will apply both Frequentist and Bayesian approaches to the existing data, as described in Section 11.4 to show the likelihood of observing more extreme values. The number of subjects evaluated by the IDMC for the two-step dose escalation to steps 2 and 3 after administration of two doses of study vaccine may continue to be 32 subjects at steps 1 and 2 as before. However, in the absence of a significant safety concern detected in the regular monitoring of all parameters of accumulating safety data, the IDMC agreed to recommend that dose escalation could potentially proceed on at least 16 subjects, without requiring the enrolment and evaluation of the full group size of 32 subjects. These amendment changes were implemented in the Synopsis and Synopsis Table 1, as well as Sections 1.2.4.1, 1.2.4.2, 1.3, Figure 1 in Section 3, Table 1 in Section 3, Sections 5.1, 6.2.2.2.1, Table 7 in 6.7.2, Table 11 in Section 6.7.3, Table 12 and Table 13 in Section 6.7.4.1, and in Sections 9.10.4 and 11.4.
- A footnote was added to indicate the requirement to record the white blood cell differential including absolute neutrophil and lymphocyte counts in the eCRF to Table 11 in Section 6.7.3 and the toxicity grading scales for the absolute neutrophil and lymphocyte counts were indicated in Table 25 of Appendix C.
- In addition, some typographical errors have been corrected throughout the protocol.

Protocol Amendment 4 Investigator Agreement

I agree:

- To conduct the study in compliance with this protocol, any future protocol amendments or protocol administrative changes, with the terms of the clinical trial agreement and with any other study conduct procedures and/or study conduct documents provided by GlaxoSmithKline (GSK) Biologicals.
- To assume responsibility for the proper conduct of the study at this site.
- That I am aware of, and will comply with, 'Good Clinical Practice' (GCP) and all applicable regulatory requirements.
- To ensure that all persons assisting me with the study are adequately informed about the GSK Biologicals' investigational vaccine(s) and other study-related duties and functions as described in the protocol.
- To acquire the reference ranges for laboratory tests performed locally and, if required by local regulations, obtain the laboratory's current certification or Quality Assurance procedure manual.
- To ensure that no clinical samples (including serum samples) are retained onsite or elsewhere without the approval of GSK Biologicals and the express written informed consent of the subject and/or the subject's legally acceptable representative.
- To perform no other biological assays on the clinical samples except those described in the protocol or its amendment(s).
- To co-operate with a representative of GSK Biologicals in the monitoring process of the study and in resolution of queries about the data.
- That I have been informed that certain regulatory authorities require the sponsor to obtain and supply, as necessary, details about the investigator's ownership interest in the sponsor or the investigational vaccine(s), and more generally about his/her financial ties with the sponsor. GSK Biologicals will use and disclose the information solely for the purpose of complying with regulatory requirements.

Hence I:

- Agree to supply GSK Biologicals with any necessary information regarding ownership interest and financial ties (including those of my spouse and dependent children).
- Agree to promptly update this information if any relevant changes occur during the course of the study and for one year following completion of the study.
- Agree that GSK Biologicals may disclose any information it has about such ownership interests and financial ties to regulatory authorities.
- Agree to provide GSK Biologicals with an updated Curriculum Vitae and other documents required by regulatory agencies for this study.

eTrack study number and Abbreviated Title	204838 (RSV PED-002)
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Date of protocol amendment	Amendment 4 Final: 10 December 2017
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Investigator name	_____
Signature	_____
Date	_____

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Sponsor Information

1. Sponsor

GlaxoSmithKline Biologicals
Rue de l'Institut, 89
1330 Rixensart, Belgium

2. Sponsor Medical Expert for the Study

Refer to the local study contact information document.

3. Sponsor Study Monitor

Refer to the local study contact information document.

4. Sponsor Study Contact for Reporting of a Serious Adverse Event

GSK Biologicals Central Back-up Study Contact for Reporting SAEs: refer to protocol Section 9.4.2.

5. GSK Biologicals' Central Safety Physician On-Call Contact information for Emergency Unblinding

GSK Biologicals Central Safety Physician and Back-up Phone contact: refer to protocol Section 9.8.

SYNOPSIS

Detailed Title	A Phase 1/2, randomized, observer-blind, controlled, multi-center, dose-escalation study to evaluate safety, reactogenicity and immunogenicity of GSK Biologicals' respiratory syncytial virus (RSV) investigational vaccine based on the RSV viral proteins F, N and M2-1 encoded by chimpanzee-derived adenovector (ChAd155-RSV) (GSK3389245A), when administered intramuscularly according to a 0, 1-month schedule to RSV-seropositive infants aged 12 to 23 months.
Indication	Active immunization of infants for the prevention of any lower respiratory tract infections (LRTI; bronchiolitis and [broncho]pneumonia) associated with respiratory syncytial virus (RSV [subtypes A and B]).
Rationale for the study and study design	Rationale for the study GlaxoSmithKline (GSK) Biologicals is developing a new investigational RSV vaccine to protect infants from RSV diseases. The candidate vaccine is based on the RSV viral proteins F, N and M2-1 encoded by chimpanzee-derived adenovector (ChAd155-RSV). The purpose of this trial is to evaluate the safety, reactogenicity and immunogenicity of this RSV candidate vaccine when first administered via intramuscular (IM) injection according to a 0, 1-month schedule to RSV-seropositive infants aged 12 to 23 months. Rationale for the study design • Study population: The immunogenicity, safety and reactogenicity of the pediatric candidate RSV vaccine (ChAd155-RSV vaccine) has been evaluated in healthy adult subjects aged 18 to 45 years (study 201974 [RSV PED-001]). The ChAd155-RSV vaccine is being developed for the active immunization of infants as from 6 weeks of age. To allow age de-escalation from the adult population to the targeted population and to not enroll and administer the ChAd155-RSV vaccine to older children who are beyond the age of severe RSV disease and would not benefit from this vaccine, the present study will be performed on RSV-seropositive infants aged 12 to 23 months, after the safety profile of the ChAd155-RSV vaccine in adults (study 201974 [RSV PED-001; NCT02491463]) was evaluated as satisfactory by an Independent Data Monitoring Committee (IDMC). The present study will provide critical information on the safety and immunogenicity profile of the ChAd155-RSV vaccine before a subsequent trial in seronegative infants.

- Control: A placebo group is included as a control for the reactogenicity, the safety and the immunogenicity assessments.

- Study blinding: Given the different storage conditions of the investigational RSV vaccine and placebo, double blinding is not possible and the study will be conducted in an observer-blind manner **(Amended 10 December 2017)**.

  When all data up to Day 60 are available, a statistical analysis will be performed. This analysis may lead to the unblinding of some subjects. As a consequence, after Day 60, the study cannot be considered as observer-blind, but will be conducted in a single-blind manner, with subjects' parent(s)/ legally acceptable representative (LAR[s]) remaining blinded up to study end (Day 730). The investigators will not have access to the treatment allocation up to study end (Day 730).

- Regimen, dose and route of administration: A vaccination regimen based on two IM injections (in the deltoid [if the muscle size is adequate; otherwise injections might be done in the anterolateral thigh]) of 5×10^9, 1.5×10^{10}, or 5×10^{10} viral particles (vp) of the ChAd155-RSV vaccine administered according to a 0, 1-month schedule (4-week interval between vaccinations) will be evaluated in this study.

  The 4-week interval regimen has been tested in pre-clinical models and results from a bovine RSV challenge model in which seronegative, colostrum-restricted newborn calves were vaccinated with two doses of 5×10^{10} vp ChAd155-RSV, four weeks apart showed that dose regimen was immunogenic and protected calves from bovine RSV disease and infection and was not associated to pulmonary pathology.

  A Phase I clinical trial evaluated a simian adenoviral vector-based RSV vaccine with the same insert (PanAd3-RSV) [RSV001 Interim Study Report, 2014]. Data from this trial, with 42 subjects, showed that a vaccination regimen based on two IM injections, separated by four weeks, was safe and immunogenic.

  Similar to the Phase I adult study (201974 [RSV PED-001]), the current study is designed in a staggered manner to allow dose escalation from 5×10^9 vp (1-log lower dose) to 5×10^{10} vp dose. A third intermediate dose of 1.5×10^{10} vp will be tested in this first time in pediatric population study to allow a more progressive escalation of the dose-related risk in this vulnerable population.

- Staggered design with 3 steps: The investigational ChAd155-RSV vaccine will be administered for the first time in a pediatric population therefore a three-step staggered design will be used to allow dose escalation of three dose levels and ensure maximum safety of the participating infants.

  The investigational ChAd155-RSV vaccine will be administered to ***up to*** 48 subjects in total. As a control, ***up to*** 48 subjects will be vaccinated with placebo across the three steps **(Amended 10 December 2017)**.

  Vaccination will be limited to a maximum of 10 infants per day. In the same investigational center, infants will be vaccinated sequentially, separated by a minimum interval of 60 minutes to allow monitoring of any acute events (e.g. hypersensitivity reaction). In addition, to monitor delayed potential reactions following vaccination, there will be a 48-hour observation period following Dose 1 and Dose 2 administration to the first eight infants (corresponding to three to five ChAd155-RSV vaccine recipients) in each Step (1, 2, and 3) before continuing vaccination of more infants (i.e. from the ninth infant). From the ninth infant, infants will be vaccinated sequentially, separated by a minimum interval of 60 minutes. Refer to the study procedures manual (SPM) for more details about the 48-hour observation period after the first eight subjects vaccinated in each Step.

  – In **Step 1**, ***between a maximum of*** 32 ***to a minimum of 16*** RSV-seropositive infants will receive two doses of either low dose ChAd155-RSV vaccine (5×10^9 vp [RSV-Ld]; 0.5 mL) or placebo (0.5 mL), according to their random assignment **(Amended 10 December 2017)**.
  – In **Step 2**, ***between a maximum of*** 32 ***to a minimum of 16*** RSV-seropositive infants will receive two doses of either middle dose ChAd155-RSV vaccine (1.5×10^{10} vp [RSV-Md]; 0.15 mL) or placebo (0.15 mL), according to their random assignment **(Amended 10 December 2017)**.
  – In **Step 3**, 32 RSV-seropositive infants will receive two doses of either high dose ChAd155-RSV vaccine (5×10^{10} vp [RSV-Hd]; 0.5 mL) or placebo (0.5 mL), according to their random assignment.

  An internal Safety Review Committee (iSRC) will review all accumulating safety data three weeks after the start of vaccination and then about every three weeks until the IDMC has reviewed all safety data up to 30 days after

administration of Dose 2 in Step 3 (i.e. Day 60).

The IDMC will review accumulating reactogenicity and safety data monthly throughout the period of vaccination (up to 30 days after administration of Dose 2 in Step 3, i.e. Day 60). The IDMC will also review incidence of all serious adverse events (SAEs) and incidence of RSV-respiratory tract infections (RTI), RSV-LRTI, RSV-severe LRTI and RSV-RTI leading to hospitalization monthly after the start of vaccination and from beginning of the first RSV season to the end of the study.

Both iSRC and IDMC will receive critical safety data (i.e. SAEs and adverse events of specific interest) within 48 hours upon GSK becoming aware of it.

The iSRC and IDMC will determine whether any of the predefined study holding rules are met. If this is the case, vaccination in the study will be immediately put on hold. At any time, the IDMC may recommend to the sponsor to suspend the enrolment to the study and/or vaccination based on the safety data regularly arising in this study.

Dose escalation will proceed after administration of the vaccine to all subjects in the previous ***step if no significant*** safety concern ***is*** detected by the IDMC in ***their*** regular ***review*** of accumulating safety data. ***The*** IDMC ***may recommend for dose escalation to proceed in the absence of a concern based on*** safety data ***review on a minimum*** of 16 subjects ***completing dose 2. The IDMC may recommend that based on the evolving safety profile, sufficient information has been accumulated to allow safe progression*** to the next ***sequential*** dose level***. Thus, the number of subjects enrolled to the low and mid dose is between 16 and 32 subjects but the number of subjects enrolled to the high dose remains fixed at 32 subjects.*** **(Amended 10 December 2017)**

Additionally, the IDMC will perform a review when all safety data up to 30 days after administration of Dose 2 in Step 3 (i.e. Day 60) are available.

Objectives

Primary

- To evaluate the safety and reactogenicity of three dose levels of the RSV investigational vaccine when administered as two IM doses according to a 0, 1-month schedule, up to 30 days after Dose 2 (i.e. Day 60) in RSV-seropositive infants aged 12 to 23 months.

Secondary

- To evaluate the safety of two IM doses of three dose levels of the RSV investigational vaccine when administered according to a 0, 1-month schedule from study start (Day 0) up to study conclusion (Day 730) in RSV-seropositive infants aged 12 to 23 months.
- To evaluate the occurrence of RSV respiratory tract infections in RSV-seropositive infants from Visit 1 (Day 0, after Dose 1) up to study conclusion (Day 730).
- To evaluate the magnitude of the CMI induced by two IM doses of three dose levels of the RSV investigational vaccine when administered according to a 0, 1-month schedule, up to Day 365 in RSV-seropositive infants aged 12 to 23 months.
- To evaluate the humoral immunogenicity induced by two IM doses of three dose levels of the RSV investigational vaccine when administered according to a 0, 1-month schedule, up to Day 365 in RSV-seropositive infants aged 12 to 23 months.

Tertiary

- To further evaluate the CMI profile induced by two IM doses of three dose levels of the RSV investigational vaccine when administered according to a 0, 1-month schedule, up to Day 365 in RSV-seropositive infants aged 12 to 23 months.
- If deemed necessary, to further characterize the immune response of the RSV investigational vaccine when two IM doses are administered according to a 0, 1-month schedule in RSV-seropositive infants.

Study design

- Experimental design: Phase I/II, observer-blind, randomized, placebo-controlled, multi-centric, study with two parallel groups per step in a 3-step staggered design.
- Duration of the study: approximately 24 months:
  - Epoch 001: primary starting at the Screening visit and ending at Visit 9 (Day 60).

– Epoch 002: follow-up starting at Visit 10 (Day 365) and ending at Visit 11 (Day 730).

Any safety, immunogenicity and disease surveillance data collected beyond Visit 9 (Day 60) will be collected in Epoch 002.

- Primary completion Date: Visit 9 (Day 60).

  End of Study: Last testing results released of samples collected at Visit 11 (Day 730).*

  * Up to Visit 11 (Day 730), there will be monthly nasal swab to detect asymptomatic RSV infections during the RSV season and if following active or a passive surveillance contacts, a subject presents symptoms of RTI, a nasal swab will be collected.

- Study groups: throughout the three steps, infants will be randomized in two groups to receive either ChAd155-RSV vaccine or placebo (Synopsis Table 1).

Synopsis Table 1 Study groups and epochs foreseen in the study

Step	Study groups	Number of subjects*	Age (Min/Max)	Epochs	
				Epoch 001	Epoch 002
1	RSV-Ld	~ 16	12 months - 23 months	x	x
	Placebo-Ld	~ 16	12 months - 23 months	x	x
2	RSV-Md	~ 16	12 months - 23 months	x	x
	Placebo-Md	~ 16	12 months - 23 months	x	x
3	RSV-Hd	~ 16	12 months - 23 months	x	x
	Placebo-Hd	~ 16	12 months - 23 months	x	x

RSV-Ld: low dose ChAd155-RSV vaccine (5 x 10^9 vp); **RSV-Md**: middle dose ChAd155-RSV vaccine (1.5 x 10^{10} vp); **RSV-Hd**: high dose ChAd155-RSV vaccine (5 x 10^{10} vp).

The target will be to enroll up to 96 RSV-seropositive infants aged 12 to 23 months at the time of first vaccination (unless the dose escalation IDMC decision can be made based on the evaluation of accumulating safety data on fewer subjects) to ensure that at least 64 infants receive two doses of study vaccine (ChAd155-RSV or Placebo). Then the number of subjects at each step will be at least 16 (approximately 8 subjects per group) at steps 1 and 2 and 32 (approximately 16 subjects per group) at step 3 **(Amended 10 December 2017)**

Synopsis Table 2 Study groups and treatment foreseen in the study

Treatment name	Volume administered	Vaccine/Product name	Study Groups					
			Step 1		Step 2		Step 3	
			RSV-Ld	Placebo-Ld	RSV-Md	Placebo-Md	RSV-Hd	Placebo-Hd
ChAd155 $5x10^9$ vp	0.5 ml	ChAd155-RSV	X					
ChAd155 $1.5x10^{10}$ vp	0.15 ml	ChAd155-RSV			X			
ChAd155 $5x10^{10}$ vp	0.5 ml	ChAd155-RSV					X	
Placebo 0.5	0.5 ml	Formulation buffer S9b		X				X
Placebo 0.15	0.15 ml	Formulation buffer S9b				X		

RSV-Ld: low dose ChAd155-RSV vaccine (5×10^9 vp); **RSV-Md**: middle dose ChAd155-RSV vaccine (1.5×10^{10} vp); **RSV-Hd**: high dose ChAd155-RSV vaccine (5×10^{10} vp).

  - Control: placebo control (Formulation buffer S9b).

- Vaccination schedule(s): two IM vaccine doses administered according to a 0, 1-month schedule (i.e. at Day 0 and Day 30).
- Treatment allocation: infants will be randomized using a randomization system on internet (SBIR) at first vaccination.
- Blinding: observer-blind in Epoch 001 and single-blind in Epoch 002 .

Synopsis Table 3 Blinding of study epochs

Study Epochs	Blinding
Epoch 001	observer-blind
Epoch 002	single-blind

- Sampling schedule:
  - Blood samples for **biochemistry** will be taken from all infants at Screening (29 days before first vaccination to Day 0) and on Day 30, and Day 60. Blood samples for **hematology** will be taken from all infants at Screening (29 days before first vaccination to Day 0) and on Day 1, Day 30, Day 31, and Day 60. Day 7 hematology testing will be performed if any ≥ Grade 1 for platelet decrease is detected on Day 1 after vaccination, to ensure its resolution. Similarly, hematology testing on Day 37 will be performed if any ≥ Grade 1 for platelet decrease is detected on Day 31. A clinical examination with special attention given to detection of spontaneous bleeding or easy bruising and evidence of bruising or petechiae will be performed by the investigator/study staff on Day 1, Day 3 and Day 7 after each vaccination (i.e. Day 1, Day 3, Day 7, Day 31, Day 33, and Day 37).

The visits on Day 3 and Day 33 may be replaced by a phone call by the investigator/study staff to inquire of the infants' general health and to solicit reports of a rash, spontaneous bleeding or easy bruising. On Day 1, and Day 31, special attention will also be given to the detection of petechiae or evidence of petechiae induced by the tourniquet. This special attention to detection of petechiae or evidence of petechiae induced by the tourniquet will also be given on Day 7 and/or Day 37 in case hematology testing will be performed on that day. Further testing may be required to investigate a finding or guide subject management based on the investigator's clinical judgment. Subjects' parent(s)/LAR(s) will be instructed to contact the investigator/study staff if their child presents symptoms of spontaneous bleeding or easy bruising or if their child develops a rash in the month following vaccination, in order to detect any thrombocytopenic petechiae or purpura. The investigator will, based on his/her medical judgement, appropriately investigate infants with clinical suspicion of low platelets.

  - Blood sample for **RSV serostatus** will be taken from all infants at Screening.
  - Blood samples for **CMI** are limited to those in investigational sites with a laboratory in proximity capable to perform WBS necessary for the CMI assay. In those sites, samples will be taken from all subjects at Screening and on Day 30, Day 60, and Day 365.
  - Blood samples for **humoral immunogenicity** will be taken from all subjects at Screening and on Day 30, Day 60, and Day 365.
  - Blood sample for **assessment of mechanism of illness (potential enhanced RSV disease [ERD])** will be taken from subjects hospitalized for LRTI (only for RSV-positive subjects using a locally available RSV test).
  - Nasal swab: there will be monthly nasal swab to detect asymptomatic RSV infections during the RSV season and if following active or a passive surveillance contacts, a subject presents symptoms of respiratory tract infections (RTI), a nasal swab will be collected.

- Study visits: Visit 1 (Day 0), Visit 5 (Day 30), Visit 9 (Day 60) and Visit 10 (Day 365) must be performed at the investigators clinical facility. Visit 2 (Day 1), Visit 4 (Day 7), Visit 6 (Day 31), Visit 8 (Day 37) and Visit 11 (Day 730) (no blood sampling for immune response and no vaccine

administration) may take place in the subject's home or at the investigators clinical facility as appropriate to the circumstances in the judgment of the investigator. Visit 3 (Day 3) and Visit 7 (Day 33) may take place at the investigators clinical facility or the investigator/clinical staff may call the subjects' parent(s)/LAR(s), as appropriate to the circumstances in the judgment of the investigator.

- Type of study: e.g. self-contained
- Data collection: Electronic Case Report Form (eCRF).
- Safety monitoring: iSRC and IDMC.
- Surveillance for RSV-RTI, difficulty in breathing and wheezing episodes.

  Surveillance period will be carried out from Visit 1 (after Dose 1) until Visit 11 (Day 730). In order to detect asymptomatic RSV-RTI, monthly nasal swabs for analysis at sponsor laboratory will be performed for all subjects during the RSV season. In order to timely detect RSV-RTI and to ensure cases are timely captured by the study sites, both active and passive surveillance will be conducted:

  - **Passive surveillance**: Parent(s)/LAR(s) are instructed to contact the investigator/study staff as soon as the subject experiences new RTI symptoms (cough, runny nose or blocked nose) or worsening of RTI symptoms, or in case of difficulty in breathing or wheezing.
  - **Active surveillance**: parent(s)/LAR(s) of all the subjects will be contacted by the investigator/study staff on a regular basis (weekly during the RSV season and every month outside the RSV season) to identify any potential RSV-RTI and to remind the parent(s)/LAR(s) of the subjects to report any new occurrence of RTI symptoms (cough, runny nose, blocked nose) or worsening of RTI symptoms, or in case of difficulty in breathing or wheezing as soon as possible.

Case definition

During the analysis of the study, all cases identified during the surveillance of RSV-RTI will be definitively classified as either RTI, LRTI, severe LRTI or very severe LRTI according to the standardized case definitions (see Synopsis Table 4) based on the available World Health Organization (WHO) case definitions.

Synopsis Table 4 Case definitions for data analysis

RSV-RTI	Runny nose OR blocked nose OR cough AND Confirmed RSV infection 4
RSV-LRTI	History of cough OR difficulty breathing 1 AND SpO2 < 95% 2, OR RR increase 3 AND Confirmed RSV infection 4
RSV-severe LRTI	Meeting the case definition of RSV-LRTI AND SpO2 < 93% 2, OR lower chest wall in-drawing
RSV-very severe LRTI	Meeting the case definition of RSV-LRTI AND SpO2 < 90% 2, OR inability to feed, OR failure to respond / unconscious
RSV hospitalization	Confirmed RSV infection 5 AND Hospitalized for acute medical condition 6
All-cause LRTI	History of cough OR difficulty breathing 1 AND SpO2 < 95% 2, OR RR increase 3

Definitions based on [Modjarrad, 2016]
LRTI = lower respiratory tract infections; RR = respiratory rate; RTI = respiratory tract infections; SpO_2 = blood oxygen saturation.
[1] Based on history reported by parents/LARs and includes difficulty breathing (e.g. showing signs of wheezing or stridor, tachypnoea, flaring [of nostrils], chest in-drawing, apnoea) associated with nasal obstruction.
[2] For blood oxygen saturation (SpO_2), the lowest value monitored will be used.
[3] RR increase defined as ≥ 40/minute (12 months of age or above).
[4] RSV infection confirmed on nasal swab positive for RSV A or B by quantitative Reverse Transcription Polymerase Chain Reaction (qRT-PCR).
[5] RSV sampling and testing is based on medical judgment of medical practitioner or driven by algorithm.
[6] Hospitalization is defined as a medical decision that the infant requires admission for observation or treatment.

Number of subjects The target will be to enroll ***up to*** 96 RSV-seropositive infants aged 12 to 23 months at the time of first vaccination ***(unless the dose escalation IDMC decision can be made based on the evaluation of accumulating safety data on fewer subjects)*** to ensure that ***at least 64*** infants receive ***two*** dose***s*** of study vaccine ***(ChAd155-RSV or Placebo)*** **(Amended 10 December 2017)**.

Endpoints

Primary

- Occurrence of AEs from first vaccination (Day 0) up to Day 60.
  - Occurrence of each solicited local and general AE, during a 7-day follow-up period after each vaccination (i.e. the day of vaccination and 6 subsequent days).
  - Occurrence of any unsolicited AE, during a 30-day follow-up period after each vaccination (i.e. the day of vaccination and 29 subsequent days).
  - Occurrence of any SAE from Day 0 up to Day 60.
  - Occurrence of episode of spontaneous or excessive bleeding (AE of specific interest), during a 30-day follow-up period after each vaccination.
  - Occurrence of any hematological (hemoglobin level, white blood cells and platelets) laboratory abnormalities at Screening, Day 1, Day 7, Day 30, Day 31, Day 37, and Day 60.
  - Occurrence of any biochemical (alanine aminotransferase, aspartate aminotransferase and creatinine) laboratory abnormalities at Screening, Day 30, and Day 60.

Secondary

- Occurrence of SAEs from study start (Day 0) up to study conclusion (Day 730).
- Occurrence of RSV-LRTI (AE of specific interest) as from Dose 1 administration up to study conclusion (Day 730).
- Occurrence of RSV-RTI, RSV-LRTI, severe RSV-LRTI (according to standardized case definitions) as from Dose 1 administration up to study conclusion (Day 730).
- Magnitude of the CMI response to the investigational RSV vaccine, pre-vaccination (Screening), post-Dose 1 (Day 30) and post-Dose 2 (Day 60 and Day 365).
  - CD3+/CD4+ T-cells expressing at least two markers among CD40L, IL-2, TNF-α, and IFN-γ upon stimulation with F, N and M2-1 peptide pools.
- Humoral response to the investigational RSV vaccine, pre-vaccination (Screening), post-Dose 1 (Day 30) and post-Dose 2 (Day 60 and Day 365):
  - Neutralizing antibody titers against RSV-A.
  - RSV F antibody concentrations.

- Humoral response to the investigational RSV vaccine, pre-vaccination (Screening), post-Dose 1 (Day 30) and post-Dose 2 (Day 60):
  - Palivizumab-competing antibody concentrations.

Tertiary

- CMI response profile (Th1, Th2, Th17) to the investigational RSV vaccine, pre-vaccination (Screening), post-Dose 1 (Day 30) and post-Dose 2 (Day 60 and Day 365).
  - CD3+/CD4+ and/or CD3+/CD8+ T-cells expressing at least one or any combination of immune marker(s) among CD40L, 41BB, IL-2, TNF-α, IFN-γ, IL-13 and IL-17 upon stimulation with F, N and M2-1 peptide pools.
- Any further exploratory immunology to detect disease-related or vaccine-related immune responses, such as but not limited to:
  - Anti-vector immunity: neutralization.

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LIST OF ABBREVIATIONS

AE:	Adverse event
ATP:	According-to-protocol
CD (CD4, CD8):	Cluster of differentiation (4, 8)
CD40L:	Cluster of differentiation 40 ligand
ChAd155:	Chimpanzee adenovirus Type 155
ChAd155-RSV:	Investigational recombinant chimpanzee adenovirus Type 155-vectored RSV vaccine
CI:	Confidence interval
CLS:	Clinical laboratory sciences
CMI:	Cell-mediated immunity
CRDL:	Clinical research & development lead
eCRF:	electronic case report form
ELISA:	Enzyme-linked immunosorbent assay
ERD:	Enhanced RSV disease
eTDF:	Electronic temperature excursion decision form
F:	RSV fusion protein
FI-RSV:	Formalin-inactivated whole virus RSV vaccine
GCP:	Good clinical practice
GMC:	Geometric mean concentration
GMT:	Geometric mean titer
GSK:	GlaxoSmithKline
HIV:	Human immunodeficiency virus
HR:	Heart rate
IB:	Investigator brochure
ICF:	Informed consent form

ICH:	International conference on harmonization
ICS:	Intracellular staining
IDMC:	Independent data monitoring committee
IEC:	Independent ethics committee
IFN-γ:	Interferon gamma
IgG :	Immunoglobulin G
IL (IL-2, IL-13, IL-17):	Interleukin (2, 13, 17)
IM:	Intramuscular
IMP:	Investigational medicinal product
IND:	Investigational New Drug
IRB:	Institutional review board
iSRC:	Internal safety review committee
LAR:	Legally acceptable representative
LRTI:	Lower respiratory tract infection
M2-1:	RSV matrix protein
MedDRA:	Medical dictionary for regulatory activities
mm³	***cubic millimeter*** **(Amended 10 December 2017)**
N:	RSV nucleocapsid protein
PCR:	Polymerase chain reaction
RNA:	Ribonucleic acid
RR:	Respiratory rate
RSV:	Respiratory syncytial virus
RSV-Hd:	High dose ChAd155-RSV vaccine (5×10^{10} vp)
RSV-Ld:	Low dose ChAd155-RSV vaccine (5×10^{9} vp)
RSV-Md:	Middle dose ChAd155-RSV vaccine (1.5×10^{10} vp)

RSV-RTI:	Respiratory tract infection associated with RSV infection
RTI:	Respiratory tract infection
RVP:	Respiratory viral panel
SAE:	Serious adverse event
SBIR:	Randomization system on internet
SPM:	Study procedures manual
SpO_2:	Blood oxygen saturation by pulse oximetry
TNF-α:	Tumor necrosis factor alpha
TVC:	Total vaccinated cohort
ULN:	Upper limit of normal
vp:	Viral particles
VSMB:	Vaccine safety monitoring board
WBS:	Whole blood stimulation
WHO:	World health organization

GLOSSARY OF TERMS

Adverse event:

Any untoward medical occurrence in a patient or clinical investigation subject, temporally associated with the use of a medicinal product, whether or not considered related to the medicinal product.

An adverse event (AE) can therefore be any unfavorable and unintended sign (including an abnormal laboratory finding), symptom, or disease (new or exacerbated) temporally associated with the use of a medicinal product. For marketed medicinal products, this also includes failure to produce expected benefits (i.e. lack of efficacy), abuse or misuse.

Blinding:

A procedure in which one or more parties to the trial are kept unaware of the treatment assignment in order to reduce the risk of biased study outcomes. The level of blinding is maintained throughout the conduct of the trial, and only when the data are cleaned to an acceptable level of quality will appropriate personnel be unblinded or when required in case of a serious adverse event. In a single-blind study, the investigator and/or his staff are aware of the treatment assignment but the subject is not. In an observer-blind study, the subject and the site and sponsor personnel involved in the clinical evaluation of the subjects are blinded while other study personnel may be aware of the treatment assignment (see Section 6.3 for details on observer-blinded studies).

Child in care:

A child who has been placed under the control or protection of an agency, organization, institution or entity by the courts, the government or a government body, acting in accordance with powers conferred on them by law or regulation. The definition of a child in care can include a child cared for by foster parents or living in a care home or institution, provided that the arrangement falls within the definition above. The definition of a child in care does not include a child who is adopted or has an appointed legal guardian.

Eligible:

Qualified for enrolment into the study based upon strict adherence to inclusion/exclusion criteria.

End of Study **(Synonym of End of Trial)**	For studies without collection of human biologicals samples or imaging data end of study is the Last Subject Last Visit (LSLV). For studies with collection of Human Biologicals Samples or imaging data, end of study is defined as the date of the last testing/reading released of the Human Biological Samples or imaging data, related to primary and secondary endpoints. End of study must be achieved no later than 8 months after LSLV
Epoch:	An epoch is a self-contained set of consecutive timepoints or a single timepoint from a single protocol. Self-contained means that data collected for all subjects at all timepoints within that epoch allows to draw a complete conclusion to define or precise the targeted label of the product. Typical examples of epochs are primary vaccinations, boosters, yearly immunogenicity follow-ups, and surveillance periods for efficacy or safety.
eTrack:	GSK's tracking tool for clinical trials.
Evaluable:	Meeting all eligibility criteria, complying with the procedures defined in the protocol, and, therefore, included in the according-to-protocol (ATP) analysis (see Sections 7.6.2 and 11.5 for details on criteria for evaluability).
Grade 1 abnormality with potential clinical relevance	Grade 1 laboratory parameters which cannot be explained or which is judged by the investigator to be potentially clinically relevant (refer to Appendix C).
Grade 1 abnormality without clinical relevance	Grade 1 laboratory parameters which can be explained by a condition which is not related to vaccination and does not increase the risk for an adverse outcome of vaccination (refer to Appendix C).
Immunological correlate of protection:	The defined immune response above which there is a high likelihood of protection in the absence of any host factors that might increase susceptibility to the infectious agent.
Infant:	As per [US Food and Drug Administration]'s definition an infant is aged one month to two years.

Investigational vaccine: **(Synonym of Investigational Medicinal Product)**	A pharmaceutical form of an active ingredient or placebo being tested or used as a reference in a clinical trial, including a product with a marketing authorization when used in a way different from the approved form, or when used for an unapproved indication, or when used to gain further information about an approved use.
Legally acceptable representative **(The terms legal representative or legally authorized representative are used in some settings.)**	An individual or juridical or other body authorized under applicable law to consent, on behalf of a prospective subject, to the subject's participation in the clinical trial.
Primary completion date:	The date that the final subject was examined or received an intervention for the purpose of final collection of data for the primary outcome, whether the clinical trial concluded according to the pre-specified protocol or was terminated.
Protocol amendment:	The International Conference on Harmonization (ICH) defines a protocol amendment as: 'A written description of a change(s) to or formal clarification of a protocol.' GSK Biologicals further details this to include a change to an approved protocol that affects the safety of subjects, scope of the investigation, study design, or scientific integrity of the study.
Protocol administrative change:	A protocol administrative change addresses changes to only logistical or administrative aspects of the study.
Randomization:	Process of random attribution of treatment to subjects in order to reduce bias of selection.
Self-contained study:	Study with objectives not linked to the data of another study.
Site Monitor:	An individual assigned by the sponsor who is responsible for assuring proper conduct of clinical studies at one or more investigational sites.
Solicited AE:	AEs to be recorded as endpoints in the clinical study. The presence/occurrence/intensity of these events is actively solicited from the subject or an observer during a specified post-vaccination follow-up period.

Subject:	Term used throughout the protocol to denote an individual who has been contacted in order to participate or participates in the clinical study, either as a recipient of the vaccine(s) or as a control.
Subject number:	A unique number identifying a subject, assigned to each subject consenting to participate in the study.
Treatment:	Term used throughout the clinical study to denote a set of investigational product(s) or marketed product(s) or placebo intended to be administered to a subject, identified by a unique number, according to the study randomization or treatment allocation.
Treatment number:	A number identifying a treatment to a subject, according to the study randomization or treatment allocation.
Unsolicited AE:	Any AE reported in addition to those solicited during the clinical study. Also any 'solicited' symptom with onset outside the specified period of follow-up for solicited symptoms will be reported as an unsolicited AE.

TRADEMARKS

The following trademarks are used in the present protocol.

Note: In the body of the protocol (including the synopsis), the names of the products will be written without the superscript symbol ™ or ® and in *italics*.

Trademarks of the GSK group of companies	Generic description
Bexsero	Meningococcal group B vaccine (recombinant, adsorbed)

Trademarks not owned by the GSK group of companies	Generic description
Synagis (MedImmune)	Recombinant humanized monoclonal anti-RSV antibodies
Allplex (Seegene)	Respiratory panel assay

1. INTRODUCTION

1.1. Background

1.1.1. RSV disease burden

Respiratory syncytial virus (RSV) is a ribonucleic acid virus of which two antigenically distinct subgroups exist, referred to as RSV-A and RSV-B [Borchers, 2013]. RSV is a highly contagious human pathogen that causes respiratory tract infections (RTI) in people of all ages.

During the first year of life, 50-70% of infants are infected with RSV and essentially all children have had an RSV infection by their second birthday. The risk for severe RSV-associated lower respiratory tract infections (LRTI) is highest in infants below 6 months of age and is a leading cause for hospitalization. It is estimated that on average 31 per 1000 children below 6 months of age are hospitalized for RSV-associated severe LRTI [Boyce, 2000; Deshpande, 2003; Hall, 2009; Holman, 2004; Iwane, 2004; Paramore, 2004; Vicente, 2003]. 50-70% of these hospitalized children do not have additional risk factors like preterm birth or cardiopulmonary disease [Boyce, 2000; García, 2010; Hall, 2009; Rietveld, 2004]. For human immunodeficiency virus (HIV)-infected children, the incidence rate of hospitalization for RSV-associated LRTI was reported to be 2.5- to 5-fold greater than in HIV-uninfected children [Cohen, 2015; Madhi, 2006]. Although RSV hospitalization rates substantially decrease after 6 months of age, a considerable number of RSV infections in children of 6-15 months of age, still lead to bronchiolitis or (broncho)pneumonia requiring medical attention [Fisher, 1997].

Previous infection with RSV does not prevent subsequent infections. Therefore, re-infection with RSV occurs throughout an individual's lifetime and is common in all age groups [Simoes, 1999; Krilov, 2011]. These re-infections generally go undiagnosed because they usually present as common acute upper respiratory tract infections. In more vulnerable persons (e.g. immunocompromized subjects or elderly), re-infections can however also lead to severe disease [Graham, 2011].

1.1.2. Current management of RSV disease in infants

To date, no vaccine is available against RSV and treatment of RSV disease is largely symptomatic and supportive care [Murray, 2014].

The antiviral drug ribavirin is currently the only approved antiviral therapy for RSV treatment, but its use is restricted to severe hospitalized cases due to uncertainties regarding its efficacy, difficulty in administration (aerosol) and high cost [American Academy of Pediatrics Subcommittee on Diagnosis and Management of Bronchiolitis, 2006].

RSV-specific monoclonal antibodies (palivizumab, *Synagis*) are indicated for use in infants at highest risk for severe RSV disease. *Synagis* is only effective as prophylaxis and is not indicated or recommended in the general, healthy infant population.

1.1.3. History of enhanced RSV disease with formalin-inactivated RSV vaccines

In the late sixties, a formalin-inactivated whole virus RSV vaccine (FI-RSV) tested in clinical trials led to more severe clinical symptoms upon subsequent natural infection with RSV in children under the age of two years. Thirty-seven out of 140 (26%) of children receiving the FI-RSV were hospitalized with severe RSV lower respiratory tract disease and there were two deaths [Polack, 2007]. The risk was greater in those children that were RSV-seronegative prior to immunization and did not occur in those that were RSV-seropositive.

This so-called 'vaccine-induced enhanced RSV disease' (ERD) is believed to have been due to the formalin inactivation step during vaccine production, leading to the induction of low-quality, non-neutralizing antibodies in RSV naïve infants [Delgado, 2004]. These antibodies did not neutralize RSV infectivity and contributed to the formation of immune complexes that may have contributed to the severe clinical symptoms and potentially immunopathology of FI-RSV ERD [Polack, 2002]. In addition, it is hypothesized that ERD may have been linked to the induction of a Th2-skewed cluster of differentiation 4 (CD4) T-cell immune response in previous naïve subjects [Castilow, 2008].

Since that time, several investigational vaccines have been explored but, despite half a century of intense RSV vaccine development, there is not a single licensed vaccine for the prevention of severe RSV disease [Anderson, 2013; Rudraraju, 2013; Murray, 2014].

1.1.4. Rationale for a viral vector-based RSV vaccine

Immunity induced by natural RSV infection is not able to fully prevent RSV re-infection, allowing re-infections to occur commonly throughout life. Both arms of the immune system are involved in protection from severe disease.

The humoral immune response is capable of neutralizing the virus and inhibiting viral replication, thereby playing a major role in protection against lower RSV infection and severe disease [Piedra, 2003]. Indeed, passive immunization with RSV-specific monoclonal antibodies (*Synagis*), when given prophylactically, has been shown to reduce RSV disease in premature infants and newborns with bronchopulmonary dysplasia or underlying cardiopulmonary disease [Cardenas, 2005].

T-cells are also involved in the control of RSV disease. Lethal RSV infections have been described in patients with low CD8 T-cell counts as in the case of severe combined immunodeficiency, bone marrow and lung transplant recipients [Hertz, 1989]. The histopathology of fatal cases of RSV infection of newborns shows that there is a relative paucity of CD8 T-cells in the lung infiltrate [Welliver, 2007]. Moreover, the presence of CD8 T-cells producing interferon gamma (IFN-γ) has been associated with diminished both Th2 responses and eosinophilia in animal models of RSV [Castilow, 2008; Stevens, 2009].

A vaccine based on recombinant viral vectors carrying relevant RSV antigens, mobilizing both humoral and cellular arms of the immune response, is considered as an adequate solution to induce a balanced and more effective immune response against the RSV virus in a naïve population. Adenoviral vector-based vaccines have been shown to be potent inducers of CD8 T-cells producing IFN-γ and antibodies against expressed antigens [Liniger, 2007; Barnes, 2012].

Potent immunogenicity and lack of prolonged transgene expression have made replication incompetent adenoviruses attractive viral vectors for vaccine development. They possess a stable virion, allowing inserts of foreign genes not to be deleted and they can infect many different cell types; the transferred information remains epichromosomal thus avoiding the risk of insertional mutagenesis.

Chimpanzee-derived adenoviruses exhibit sequence homology to human adenoviruses within the hexon protein in particular, which is a major capsid protein used for subgroup classification of adenoviruses [Roy, 2011; Colloca, 2012]. They are not known to cause pathological illness in humans and antibodies against chimpanzee adenoviruses have low or no seroprevalence ranging from 0-4% in Europe and in the United States, up to 20% in the developing countries, which is far less than the most common human adenovirus serotype 5 (Ad5), with seroprevalence rates of 40–45% in the United States and up to 90% in sub-Saharan Africa [Capone, 2013].

In conclusion, the chimpanzee-derived adenovectors have the following important features:

- Weak neutralization by human sera.
- Potent immunogenicity, mainly Th1-directed.
- Track record of good tolerability in human clinical trials.

1.1.5. Antigens selected for the vaccine

The investigational vaccine is a recombinant viral vector manufactured using a synthetic deoxyribonucleic acid fragment that encodes three RSV proteins:

- The fusion (F) protein deleted from the transmembrane and cytoplasmic regions (F0ΔTM). The F protein is a major surface antigen of the RSV virus that is well conserved among RSV-A and RSV-B subgroups. In addition, it is the main target of the neutralizing antibody response to RSV, which is considered essential for protection against RSV-associated severe disease [Magro, 2012].
- The nucleocapsid (N) protein is an internal (non-exposed) antigen, highly conserved between RSV strains and known to be a source of many T-cell epitopes [Anderson, 2010; Townsend, 1984]. The N protein is essential for the replication and transcription of the RSV genome. Its primary function is to encapsidate the virus genome protecting it from ribonucleases.
- The matrix (M2-1) protein is a transcription anti-termination factor that is important for the efficient synthesis of full-length messenger ribonucleic acid (RNA) as well as for the synthesis of polycistronic readthrough messenger RNA, which are

characteristic of non-segmented negative-strand RNA viruses. M2-1 is an internal (non-exposed) antigen, highly conserved between RSV strains and known to be a source of many T-cell epitopes [Anderson, 2010; Townsend, 1984].

The N and M2-1 antigens were therefore included in the vaccine construct as source of T-cell epitopes for the induction of cell-mediated immunity (CMI).

Please refer to the current Investigator Brochure (IB) for additional information.

1.1.6. Pre-clinical and clinical experience

The candidate vaccine is based on the RSV viral proteins F, N and M2-1 encoded by a chimpanzee-derived adenovector (ChAd155-RSV). A number of pre-clinical studies were conducted and have demonstrated immunogenicity, efficacy and safety of intramuscular (IM) injection of the ChAd155-RSV vaccine. Several animal models have been selected for such studies: mice, cotton rats and newborn calves. Importantly, vaccination has not induced hallmarks of enhanced pathology in these models.

The immunogenicity, safety and reactogenicity of the pediatric candidate RSV vaccine (ChAd155-RSV vaccine) have been evaluated in healthy adults aged 18 to 45 years (study 201974 [RSV PED-001; NCT02491463]) There were no significant safety concerns identified up to Day 60 in study RSV PED-001. Overall, the ChAd155-RSV vaccine high dose (Hd) (5×10^{10} vp) seems to be more reactogenic (local and general) than the ChAd155-RSV vaccine low dose (Ld) (5×10^{9} vp), however, the reactogenicity profile was less than that observed in the Bexsero group. No safety signal from the assessed hematology parameters (hemoglobin, platelet count, prothrombin time and APTT) was observed in subjects receiving the ChAd155-RSV vaccine. No significant reductions in platelet count or clinically significant changes in coagulation parameters were observed up to 30 days post Dose 2. An approximately 2.4-fold increase in RSV-A neutralizing antibody titers (geometric mean titer [GMT] from baseline) was observed in both RSV-Ld and RSV-Hd after Dose 1. No booster effect was evident after Dose 2. An anti-F IgA and IgG antibody secreting B-cells response and an RSV F, N and M2-1 specific IFN-γ secreting T-cells response in RSV-Hd group after the first dose were observed with ELISpot. There was no booster response after the second vaccination. There was no specific vaccine-induced CD4 T-cellular response observed with intracellular staining (ICS). For CD8 T-cells only a weak CD8 IFN-γ response to N was shown with ICS for some subjects.

Please refer to the current IB for information regarding the pre-clinical and clinical studies of GlaxoSmithKline (GSK) Biologicals' investigational ChAd155-RSV vaccine.

1.2. Rationale for the study and study design

1.2.1. Rationale for the study

GSK Biologicals is developing the pediatric candidate RSV vaccine (ChAd155-RSV vaccine) to protect infants from RSV diseases.

The purpose of this trial is to evaluate the safety, reactogenicity and immunogenicity of the ChAd155-RSV vaccine when first administered via IM injection according to a 0, 1-month schedule to RSV-seropositive infants aged 12 to 23 months.

1.2.2. Rationale for the choice of study population

1.2.2.1. Study population

The immunogenicity, safety and reactogenicity of the pediatric candidate RSV vaccine (ChAd155-RSV vaccine) has been evaluated in healthy adults aged 18 to 45 years (study 201974 [RSV PED-001; NCT02491463]). The ChAd155-RSV vaccine is being developed for the active immunization of infants as from 6 weeks of age. To allow age de-escalation from the adult population to the targeted population and to not enroll and administer the ChAd155-RSV vaccine to older children who are beyond the age of severe RSV disease and would not benefit from this vaccine, the present study will be performed on RSV-seropositive infants aged 12 to 23 months, after the safety profile of the ChAd155-RSV vaccine in adults (study 201974 [RSV PED-001; NCT02491463]) was evaluated as satisfactory by an Independent Data Monitoring Committee (IDMC). The present study will provide critical information on the safety and immunogenicity profile of the ChAd155-RSV vaccine before a subsequent trial in seronegative infants.

1.2.2.2. RSV serostatus using IBL International kit and choice of study population

To decrease the risk of ERD (refer to Section 1.1.3) in the present study, RSV-seropositive infants aged 12 to 23 months will be enrolled.

RSV-seropositive infants will be screened using an enzyme-linked immunosorbent assay (ELISA) method (IBL International kit), detecting immunoglobulin G (IgG) antibody against RSV F or G protein. The objective of the screening for RSV-seropositive infants is to limit the evaluation of the ChAd155-RSV vaccine to infants previously primed by RSV and therefore considered at low risk for ERD. However this assay does not discriminate between maternal antibodies and those naturally acquired by exposure to RSV. For this reason, although the ChAd155-RSV vaccine is not a FI-RSV vaccine, risk of ERD might still exist while vaccinating seropositive infants that have never been naturally exposed to RSV (RSV-naïve seropositive infants). However, GSK considers the risk of enrolling RSV-seronegative (i.e. false RSV-seropositive) infants in the present study as extremely low, for the two following reasons:

- Beyond 12 months of age the number of RSV-naïve seropositive infants is expected to be very low. Indeed, published data suggest that the RSV seropositivity of infants aged 12 to 23 months is rather due to natural exposure to RSV than to maternal antibodies. In a randomized clinical trial performed in Bangladesh in which infants' serum samples were collected at birth, 6, 10, 16, 20, 24, and 72 weeks of age the RSV-neutralizing antibody half-life in infants was 38 days and decreased below a protective titer by a median of 17 weeks. This corresponds to the clinical observation that infants are most vulnerable to developing severe RSV disease between 6 weeks and 6 months of age, a period when maternal antibodies are waning and the infant immune response may be incapable of generating a robust neutralizing antibody

response to infection [Chu, 2014]. In another study, a birth-cohort study performed in Kenya, the maternal-RSV antibodies in serum measured by ELISA declined rapidly following birth (half-life: 79 days; [Ochola, 2009]). Another birth-cohort study performed in Kenya, showed also a rapid decay in RSV-neutralizing antibodies, with an half-life of 36 days [Nyiro, 2015].

- Despite the assay characterization performed by the manufacturer, a potential lack of sensitivity of the IBL International kit compared to a RSV neutralization assay has been reported [Malkin, 2013]. GSK considers that the specificity and sensitivity characteristics of the assay indicate a reduced risk of enrolling RSV-seronegative infants identified RSV-seropositive in the assay (i.e. false positive results).

Given these facts and data it is unlikely that infants aged 12 months or above reported RSV-seropositive by the IBL International kit will be RSV-seronegative and it was decided to perform this study in infants aged 12 to 23 months.

1.2.3. Rationale for regimen, dose and route of administration

A vaccination regimen based on two IM injections (in the deltoid [if the muscle size is adequate; otherwise injections might be done in the anterolateral thigh]) of 5 x 10^9, 1.5 x 10^{10}, or 5 x 10^{10} viral particles (vp) of the ChAd155-RSV vaccine administered according to a 0, 1-month schedule (4-week interval between vaccinations) will be evaluated in this study.

The 4-week interval regimen has been tested in pre-clinical models and results from a bovine RSV challenge model in which seronegative, colostrum-restricted newborn calves were vaccinated with two doses of 5 x 10^{10} vp ChAd155-RSV, four weeks apart showed that dose regimen was immunogenic and protected calves from bovine RSV disease and infection and was not associated to pulmonary pathology.

Using a different adenovector, PanAd3, the 5 x 10^{10} vp dose regimen was shown to be safe in toxicology studies in mice. Pre-clinical data in different species (mice, cotton rats, non-human primates, and seronegative newborn calves) have shown that this regimen induces both anti-RSV F neutralizing antibodies and systemic T-cell responses [Taylor, 2015; Pierantoni, 2015].

A Phase I clinical trial evaluated a simian adenoviral vector-based RSV vaccine with the same insert (PanAd3-RSV) [RSV001 Interim Study Report, 2014]. Data from this trial, with 42 subjects, showed that a vaccination regimen based on two IM injections, separated by four weeks, was safe and immunogenic.

Similar to the Phase I adult study (201974 [RSV PED-001]), the current study is designed in a staggered manner to allow dose escalation from 5 x 10^9 vp (1-log lower dose) to 5 x 10^{10} vp dose. A third intermediate dose of 1.5 x 10^{10} vp will be tested in this first time in pediatric population study to allow a more progressive escalation of the dose-related risk in this vulnerable population.

Please refer to the current IB for information regarding the pre-clinical, clinical and toxicology studies of GSK Biologicals' investigational ChAd155-RSV vaccine.

1.2.4. Rationale for safety monitoring plan

1.2.4.1. *Rationale for earlier dose escalation decision making*

Under Amendment 4 of the protocol, the IDMC may on reviewing of the accumulating safety data recommend that dose escalation can safely proceed based on the experience of a minimum of 16 subjects.

Taking the a priori safety concern of thrombocytopenia, both Frequentist and Bayesian approaches are applied to provide complementary assurance on the possibility to exclude a signal based on the accumulated safety data. The Frequentist approach examines the statistical power to rule out a significant decrease in platelet counts due to vaccination that would suggest a four-fold or greater increase in the risk of a Grade 3 or higher thrombocytopenia with 16 subjects per dose cohort. The Bayesian approach calculates the posterior predictive probability of not observing any subject with a Grade 3 or higher thrombocytopenia among the future 16 subjects by basing it on what we have observed in the first 16 subjects (see Section 11.4). **(Amended 10 December 2017)**

1.2.4.2. Staggered design with 3 steps

The investigational ChAd155-RSV vaccine will be administered for the first time in a pediatric population therefore a three-step staggered design will be used to allow dose escalation of three dose levels and ensure maximum safety of the participating infants.

The target will be to enroll ***up to*** 96 RSV-seropositive infants ***aged 12 to 23 months at the time of first vaccination (unless the dose escalation IDMC decision can be made based on the evaluation of accumulating safety data on fewer subjects)*** to ensure that ***at least 64*** infants receive ***two*** dose***s*** of study vaccine ***(ChAd155-RSV or Placebo)***. **(Amended 10 December 2017)**.

Vaccination will be limited to a maximum of 10 infants per day. In the same investigational center, infants will be vaccinated sequentially, separated by a minimum interval of 60 minutes to allow monitoring of any acute events (e.g. hypersensitivity reaction). In addition, to monitor delayed potential reactions following vaccination, there will be a 48-hour observation period following Dose 1 and Dose 2 administration to the first eight infants (corresponding to three to five ChAd155-RSV vaccine recipients) in each Step (1, 2, and 3) before continuing vaccination of more infants (i.e. from the ninth infant). From the ninth infant, infants will be vaccinated sequentially, separated by a minimum interval of 60 minutes. Refer to the study procedures manual (SPM) for more details about the 48-hour observation period after the first eight subjects vaccinated in each Step.

- In **Step 1**, ***between a maximum of*** 32 ***to a minimum of 16*** RSV-seropositive infants will receive two doses of either low dose ChAd155-RSV vaccine (5×10^9 vp [RSV-Ld]; 0.5 mL) or placebo (0.5 mL), according to their random assignment **(Amended 10 December 2017)**.

- In **Step 2**, ***between a maximum of*** 32 ***to a minimum of 16*** RSV-seropositive infants will receive two doses of either middle dose ChAd155-RSV vaccine (1.5×10^{10} vp [RSV-Md]; 0.15 mL) or placebo (0.15 mL), according to their random assignment **(Amended 10 December 2017**.
- In **Step 3,** 32 RSV-seropositive infants will receive two doses of either high dose ChAd155-RSV vaccine (5×10^{10} vp [RSV-Hd]; 0.5 mL) or placebo (0.5 mL), according to their random assignment.

An internal Safety Review Committee (iSRC) will review all accumulating safety data three weeks after the start of vaccination and then about every three weeks until the IDMC has reviewed all safety data up to 30 days after administration of Dose 2 in Step 3 (i.e. Day 60). Ad-hoc iSRC meetings may be held should safety concerns warrant a safety review. Refer to Section 9.10.2 for more information about iSRC.

The IDMC will review accumulating reactogenicity and safety data monthly throughout the period of vaccination (up to 30 days after administration of Dose 2 in Step 3, i.e. Day 60). The IDMC will also review incidence of all serious adverse events (SAEs) and incidence of RSV-respiratory tract infections (RTI), RSV-LRTI, RSV-severe LRTI and RSV-RTI leading to hospitalization monthly after the start of vaccination and from beginning of the first RSV season to the end of the study.

Both iSRC and IDMC will receive critical safety data (i.e. SAEs and adverse events of specific interest) within 48 hours upon GSK becoming aware of it.

The iSRC and IDMC will determine whether any of the predefined study holding rules are met. If this is the case, vaccination in the study will be immediately put on hold. At any time, the IDMC may recommend to the sponsor to suspend the enrolment to the study and/or vaccination based on the safety data regularly arising in this study.

Dose escalation will proceed after administration of ***two doses of*** the vaccine to 32 subjects. ***However,*** in the absence of a ***significant*** safety concern detected in the regular monitoring of accumulating safety data ***on at least 16 subjects, the IDMC may allow for dose escalation to proceed without the requirement of enrolling and evaluating the full group size of 32 subjects. A subsequent dose escalation will proceed after administration of two doses of the vaccine to between a maximum of 32 to a minimum of 16 subjects in in the absence of a significant safety concern detected by the IDMC in the regular monitoring of accumulating safety data. Thus, upon review of the accumulating safety data on a minimum of 16 subjects after administration of two doses of the vaccine, the IDMC may decide that based on the evolving safety profile, sufficient information has been accumulated to allow safe progression to the next sequential dose level*** **(Amended 10 December 2017).** It will be ensured that, as a minimum the IDMC will have reviewed safety data of at least 16 subjects for two doses and given their formal approval prior to enrolling subjects to the next dose level.

Additionally, the IDMC will perform a review when all safety data up to 30 days after administration of Dose 2 in Step 3 (i.e. Day 60) are available.

Refer to Section 9.10.3 for more information about IDMC.

1.2.4.3. Rationale for monitoring of hematology parameters

Emphasis on "minimal risk" is imperative for the protection of the pediatric population participating to research studies where blood sample volume limits is a critical parameter. To stay within blood sampling limits that are consistent with physiological "minimal risk" in this young and vulnerable population, but also to minimize any development of needle aversion, blood draws for the determination of hematological parameters are scheduled at Day 1 and Day 31 (corresponding to the first day after administration of the vaccine). Additional specimens will be drawn if any ≥ Grade 1 for platelet decrease is detected at Day 1 or Day 31 to investigate clinical symptoms suggestive of low platelets. Blood samples collected within the screening window will serve as baseline for the immunological analysis and for the hematology and biochemistry check-up before vaccination. Consequently, the maximum blood draw volumes collected per protocol have been restricted to 9.0 mL within 24 hours and 26.2 mL over eight weeks (refer to Table 5 and Table 7). For investigational sites without a laboratory in proximity capable to perform whole blood stimulation [WBS] necessary for the CMI assay the volumes will be less. Of note, it is considered that blood sample volumes ranging from 1 to 5% of total blood volume within 24 hours and up to 10% of total blood volume over 8 weeks are consistent with the limited evidence available on the minimal physiological risk for healthy children [Howie; 2011]. The total blood volume of a child is around 75 to 80 mL/kg (85 to 105 mL/kg in the neonatal period). Considering a girl of 12 months of age with a low weight of 7.3 kg (5th percentile [WHO; 2006]), the minimal individual total blood volume in the study population would be 547.5 mL and therefore the safer limits of blood draw volumes would range from 5.5 to 27.4 mL within 24 hours and be limited to 54.7 mL over 8 weeks. To minimize any development of needle aversion, pain relief by means of a topical local anesthetic may be offered to infants prior to any blood sampling requested by the protocol at the discretion of the investigator.

During a study carried out in adult subjects (RSV001), a mild non-clinically significant drop in hemoglobin was noted following vaccination with another adenoviral vector (PanAd3-RSV) without clinical signs and with a reversal towards baseline values over time [RSV001 Interim Study Report, 2014]. In the repeat dose toxicology study in rabbits using the ChAd155-RSV vaccine, a transient non-clinically significant drop in platelets was noted post IM vaccination (maximal drop of platelet observed 24 hours after vaccination; refer to the current IB for further details). In light of these data, a clinical examination with special attention given to the detection of spontaneous bleeding or easy bruising and evidence of bruising or petechiae will be performed by the investigator/study staff on Day 1, Day 3, Day 7, Day 31, Day 33 and Day 37. The visits on Day 3 and Day 33 may be replaced by a phone call by the investigator/study staff to inquire of the infants' general health and to solicit reports of a rash, spontaneous bleeding or easy bruising. On Day 1 after each vaccination, special attention will also be given to the detection of petechiae or evidence of petechiae induced by the tourniquet. This special attention to detection of petechiae or evidence of petechiae induced by the tourniquet will also be given on Day 7 and Day 37 in case hematology testing will be performed on that day. Subjects' parent(s)/ legally acceptable representative(s) (LAR[s]) will be instructed to contact the investigator/study staff if their child presents symptoms of spontaneous bleeding or easy bruising or if their child develops a rash in the month following vaccination, in order to detect any thrombocytopenic petechiae or purpura. The

investigator will, based on his/her medical judgement, appropriately investigate infants with clinical suspicion of low platelets.

Note that a broad safety hematology and biochemistry evaluation was performed at each study visit in adults during study RSV PED-001 (201974; NCT02491463). In this study, blood samples for hematology/biochemistry were taken from all subjects at all study visits, i.e. at Screening, on the day of vaccination (Day 0 and Day 30), 1 day post vaccination (Day 1 and Day 31), 3 days post vaccination (Day 3 and 33), 7 days post vaccination (Day 7 and 37), 30 days post Dose 2 (Day 60), on Day 180 and on Day 360. The safety profile of the ChAd155-RSV vaccine was found to be satisfactory by an IDMC. No safety signal from the assessed hematology parameters (hemoglobin, platelet count, prothrombin time and APTT) was observed in subjects receiving the ChAd155-RSV vaccine. No significant reductions in platelet count or clinically significant changes in coagulation parameters were observed up to 30 days post Dose 2 (refer to Section 1.1.6 and current IB).

1.2.4.4. Rationale for RSV disease surveillance

Although the risk of ERD is thought to be very low with an adenovirus-based vaccine (refer to Section 1.3.1), all children will be closely monitored by active and passive surveillance involving a clinical visit within 24 hours based on any suspicion of difficulty in breathing, wheezing or parental concern associated with symptoms of an RTI (refer to Section 9.2). This will ensure prompt assessment of need for medical care. Furthermore, a limited number of subjects will be enrolled in this study. All of them will be under surveillance allowing description of RSV associated LRTI and severe LRTI incidence in the ChAd155-RSV vaccine groups and the Placebo groups. All subjects experiencing LRTI associated with RSV will be considered as adverse events (AEs) of specific interest (refer to Section 9.1.5.2) and will have to be reported within 24 hours. In order to support this timely reporting, the investigator will make the diagnosis according to his/her medical judgment and locally-available diagnostic tests of RSV infections. Final analysis of RSV-LRTI will be performed according to standardized case definitions for clinical symptomatology and diagnostic test at sponsor laboratory (refer to Table 4) ensuring consistency across sites.

1.2.5. Rationale for study blinding

Given the different storage conditions of the investigational RSV vaccine and placebo, double blinding is not possible and the study will be conducted in an observer-blind manner **Amended 10 December 2017)**.

When all data up to Day 60 are available, a statistical analysis will be performed. This analysis may lead to the unblinding of some subjects. As a consequence, after Day 60, the study cannot be considered as observer-blind, but will be conducted in a single-blind manner, with subjects' parent(s)/ LAR(s) remaining blinded up to study end (Day 730). The investigators will not have access to the treatment allocation up to study end (Day 730).

Please refer to the glossary of terms for the definition of observer-blind and single-blind.

1.2.6. Rationale for the use of placebo

The placebo group is included as a control for the reactogenicity, the safety and the immunogenicity assessments.

1.3. Benefit : Risk Assessment

Approximately half of the infants (***between 32 and*** 48) in this study will be exposed to the ChAd155-RSV vaccine, whereas the other infants will receive a placebo **(Amended 10 December 2017)**.

Please refer to the current IB for the summary of potential risks and benefits of ChAd155-RSV vaccine.

The following section outlines the risk assessment and mitigation strategy for this study protocol:

1.3.1. Risk Assessment

As with all injectable vaccines, immediate systemic allergic reactions to vaccination can occur. In order to be able to treat infants with an immediate systemic allergic reaction to vaccination, all infants across all steps will need to remain under observation (visual follow-up as well as measurement of vital signs) at the study site for at least 60 minutes after vaccination.

Syncope (fainting) can occur following or even before any vaccination as a psychogenic response to the needle injection. Therefore, it is important that procedures are in place to avoid injury from fainting.

Risks linked to the investigational RSV vaccine

This study is conducted in RSV-seropositive infants who are assumed to have been previously infected with RSV and therefore not at risk for ERD. The detection of antibody does not discriminate between maternally acquired antibodies and those naturally acquired by exposure to RSV. However, the risk of misclassification is believed to be low in infants from 6 months of age since in published data the estimated half-life of maternally acquired RSV antibody ranged from 31 to 79 days [Chu, 2014; Ochola, 2009; Nyiro, 2015].

IM vaccination commonly precipitates a transient and self-limiting local inflammatory reaction. This may typically include redness, swelling and tenderness.

Most systemic symptoms observed in a clinical trial with a similar product carried out in healthy adults (chimpanzee adenovirus-based vaccine with ChAd3 as vector for vaccination against hepatitis C) at doses to be used in this trial did not exceed mild severity [HCV001 Clinical Study Report, 2011]. Fatigue, headache and malaise were the most commonly reported systemic AEs overall.

In a Phase I clinical trial with a similar simian adenoviral vector-based RSV vaccine with the same insert (PanAd3-RSV) carried out in healthy adults, the most commonly reported AEs concerned injection site reactions. However, a mild non-clinically significant drop in hemoglobin was noted following vaccination without clinical signs and with a reversal towards baseline values over time [RSV001 Interim Study Report, 2014].

A transient non-clinically significant drop in platelets was noted post IM vaccination in a preclinical study with the ChAd155-vector (refer to the current IB for further details). Furthermore, in Ebola Phase I studies in adults investigating a similar adenoviral vectored vaccine (ChAd3-EBO-Z), transient decreases in thrombocyte counts were also observed. These decreases occurred mostly on Day 1 after vaccination and generally returned to baseline by Day 7. Although most of these decreases remained within the normal range, the per protocol criteria for thrombocytopenia (i.e. thrombocyte count of < 150 x 10^3/µL) were met for 2.6% (7 out of 270) of the vaccinated subjects. None of the decreases in thrombocyte counts or the cases of thrombocytopenia were clinically significant (i.e. no clinical signs or symptoms suggestive of increased tendency to bleeding were reported in any of the subjects; refer to the current IB of recombinant chimpanzee adenovirus Type 3-vectored Ebola Zaire vaccine for further details). Although the mechanism underlying these decreases currently remains unclear, it is well described in literature that, post intravenous administration, adenovirus activates platelets and induces platelet-leukocyte aggregate formation, causing an associated increase in platelet and leukocyte-derived microparticles [Othman, 2007; Stone, 2007].

No significant safety concerns were identified up to 30 days post Dose 2 in study RSV PED-001 with the ChAd155-RSV vaccine in healthy adults aged 18 to 45 years (refer to Section 1.1.6 and to the current IB).

All available safety information from clinical and pre-clinical studies was evaluated by GSK's Safety Review Team. It was concluded that the available information did not pose an immediate safety concern, since the decreases seen were transient and not clinically significant. Nevertheless, since this is the first time the ChAd155-RSV vaccine is administered to infants and to ensure a maximum safety follow-up, a safety monitoring plan has been put in place (refer to Section 1.2.4).

1.3.2. Benefit Assessment

The infants receiving the investigational ChAd155-RSV vaccine may not directly benefit from vaccination since the efficacy of the investigational ChAd155-RSV vaccine has not been assessed yet and it is hence not known whether it is effective in protecting against RSV infection.

Subjects' parent(s)/ LAR(s) could gain medical advices about their infant's general health status through the medical evaluations/assessments associated with this study (i.e. physical examination, blood testing [hematology and biochemistry data], surveillance for RTI and LRTI).

1.3.3. Overall Benefit:Risk Conclusion

The investigational ChAd155-RSV vaccine is currently in a very early stage of clinical development and no vaccine efficacy has been demonstrated. Taking into account the measures taken to minimize the risk to infants participating in this study, the potential risks to the subjects are justified by the potential benefits linked to the development of this pediatric RSV vaccine.

2. OBJECTIVES

2.1. Primary objective

- To evaluate the safety and reactogenicity of three dose levels of the RSV investigational vaccine when administered as two IM doses according to a 0, 1-month schedule, up to 30 days after Dose 2 (i.e. Day 60) in RSV-seropositive infants aged 12 to 23 months.

Refer to Section 11.1 for the definition of the primary endpoints.

2.2. Secondary objectives

- To evaluate the safety of two IM doses of three dose levels of the RSV investigational vaccine when administered according to a 0, 1-month schedule from study start (Day 0) up to study conclusion (Day 730) in RSV-seropositive infants aged 12 to 23 months.
- To evaluate the occurrence of RSV respiratory tract infections in RSV-seropositive infants from Visit 1 (Day 0, after Dose 1) up to study conclusion (Day 730).
- To evaluate the magnitude of the CMI induced by two IM doses of three dose levels of the RSV investigational vaccine when administered according to a 0, 1-month schedule, up to Day 365 in RSV-seropositive infants aged 12 to 23 months.
- To evaluate the humoral immunogenicity induced by two IM doses of three dose levels of the RSV investigational vaccine when administered according to a 0, 1-month schedule, up to Day 365 in RSV-seropositive infants aged 12 to 23 months.

Refer to Section 11.2 for the definition of the secondary endpoints.

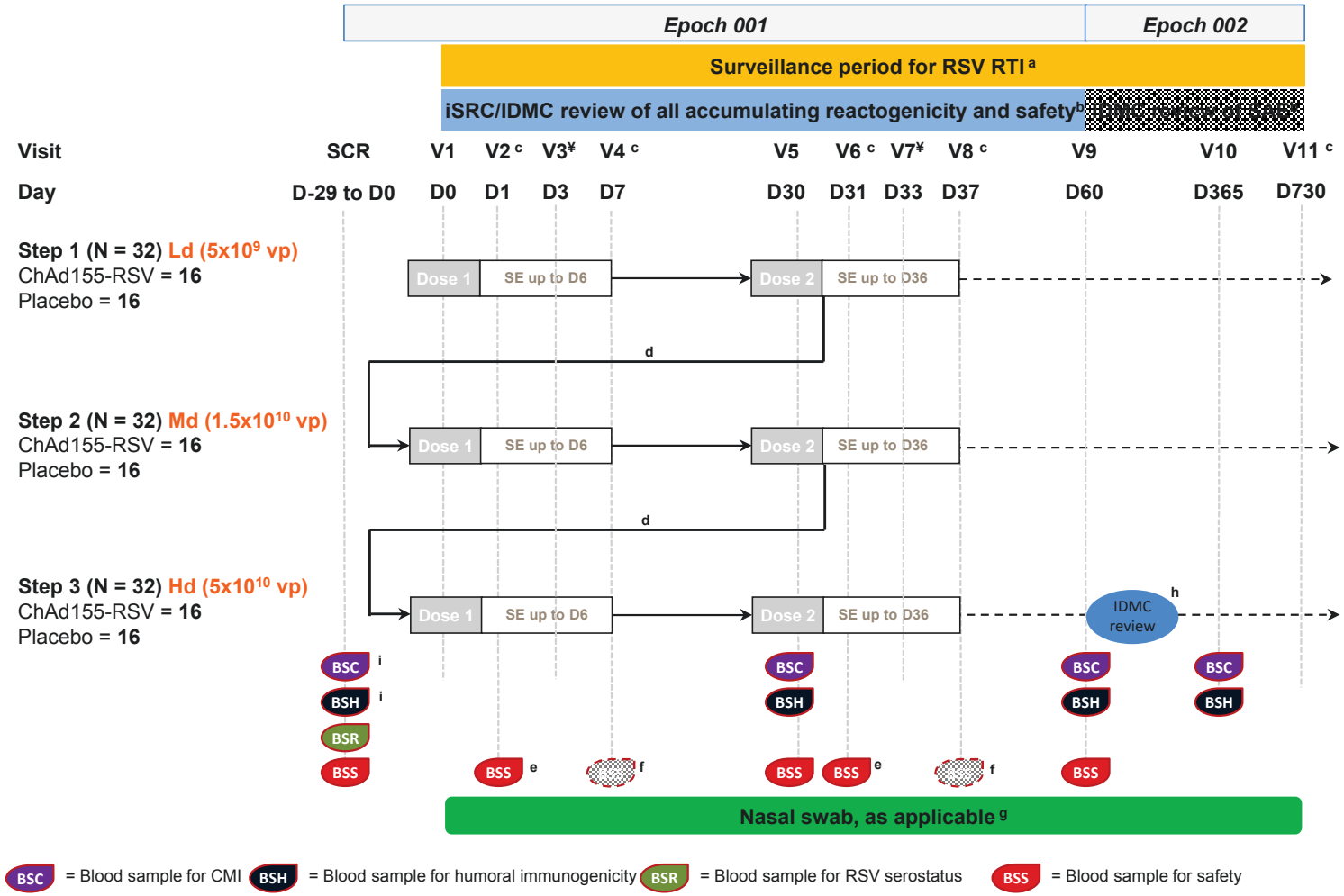
2.3. Tertiary objectives

- To further evaluate the CMI profile induced by two IM doses of three dose levels of the RSV investigational vaccine when administered according to a 0, 1-month schedule, up to Day 365 in RSV-seropositive infants aged 12 to 23 months.
- If deemed necessary, to further characterize the immune response of the RSV investigational vaccine when two IM doses are administered according to a 0, 1-month schedule in RSV-seropositive infants

Refer to Section 11.3 for the definition of the tertiary endpoints.

3. STUDY DESIGN OVERVIEW

Figure 1 Study design



Hd: High dose; **IDMC:** Independent Data Monitoring Committee; **iSRC:** Internal Safety Review Committee; **Ld:** low dose; **Md:** middle dose; **RSV:** respiratory syncytial virus; **SCR:** Screening; **SE:** solicited events; **vp:** viral particles.

¥ On Day 3 and Day 33, a visit may take place at the investigators clinical facility or the investigator/clinical staff may call the subjects' parent(s)/LAR(s), as appropriate to the circumstances in the judgment of the investigator.

a Surveillance for RSV-RTI comprises monthly nasal swab collected to detect asymptomatic RSV infections during RSV season and active and passive surveillance contacts for RSV symptomatic RTI (see Section 9.2). Data about RSV–RTI incidence will be reviewed monthly by an IDMC.

b Within each step, an iSRC will review all accumulating safety data three weeks after the start of vaccination and then about every three weeks (until the IDMC has reviewed all safety data up to 30 days after administration of Dose 2 in Step 3 i.e. Day 60). The IDMC will review all accumulating reactogenicity and safety data monthly throughout the period of vaccination (up to 30 days after administration of Dose 2 in Step 3, i.e. Day 60) and accumulating SAEs until Day 730. Refer to Sections 9.10.2 and 9.10.3.

c Visit 2 (Day 1), Visit 4 (Day 7), Visit 6 (Day 31), Visit 8 (Day 37) and Visit 11 (Day 730) may take place in the subject's home or at the investigators clinical facility as appropriate to the circumstances in the judgment of the investigator.

d Dose escalation will proceed after administration of ***two doses of*** the vaccine to 32 subjects in Step ***1***. ***However,*** in the absence of a ***significant*** safety concern detected in the regular monitoring of accumulating safety data ***on at least 16 subjects, the IDMC may allow for dose escalation to proceed without the requirement of enrolling and evaluating the full group size of 32 subjects. A subsequent dose escalation (from Step 2 to Step 3) will proceed after administration of two doses of the vaccine to between a maximum of 32 to a minimum of 16 subjects in the absence of a significant safety concern detected by the IDMC in the regular monitoring of accumulating safety data. Thus, upon review of the accumulating safety data on a minimum of 16 subjects after administration of two doses of the vaccine, the IDMC may decide that based on the evolving safety profile, sufficient information has been accumulated to allow safe progression to the next sequential dose level***. **(Amended 10 December 2017)** It will be ensured that, as a minimum the IDMC will have reviewed safety data of at least 16 subjects for two doses and given their formal approval prior to enrolling subjects to the next dose level.

e Only for hematology.

f Day 7 hematology testing will be performed if any ≥ Grade 1 for platelet decrease is detected on Day 1 after vaccination; to ensure its resolution. Similarly, hematology testing on Day 37 will be performed if any ≥ Grade 1 for platelet decrease is detected on Day 31.

g Refer to Section 6.6.11.3.

h The IDMC will perform a review when all safety data up to 30 days after administration of Dose 2 in Step 3 (i.e. Day 60) are available.

i Blood samples for CMI and for humoral immunogenicity may be postponed until after the RSV serostatus is known and eligibility is confirmed up to or on Day 0 (but before vaccination).

Protocol waivers or exemptions are not allowed with the exception of immediate safety concerns. Therefore, adherence to the study design requirements, including those specified in the outline of study procedures (Section 6.5), are essential and required for study conduct.

- Experimental design: Phase I/II, observer-blind, randomized, placebo-controlled, multi-centric, study with two parallel groups per step in a 3-step staggered design.
- Duration of the study: approximately 24 months:
  - Epoch 001: primary starting at the Screening visit and ending at Visit 9 (Day 60).
  - Epoch 002: follow-up starting at Visit 10 (Day 365) and ending at Visit 11 (Day 730).

  Any safety, immunogenicity and disease surveillance data collected beyond Visit 9 (Day 60) will be collected in Epoch 002.
- Primary completion Date: Visit 9 (Day 60).

  Refer to glossary of terms for the definition of primary completion date.
- End of Study: Last testing results released of samples collected at Visit 11 (Day 730).*

  * Up to Visit 11 (Day 730), there will be monthly nasal swab to detect asymptomatic RSV infections during the RSV season and if following active or a passive surveillance contacts, a subject presents symptoms of RTI, a nasal swab will be collected.

  Refer to glossary of terms for the definition of end of study.
- Study groups: throughout the three steps, infants will be randomized in two groups to receive either ChAd155-RSV vaccine or placebo (Table 1).

Table 1 Study groups and epochs foreseen in the study (Amended 10 December 2017)

Step	Study groups	Number of subjects*	Age (Min/Max)	Epochs	
				Epoch 001	Epoch 002
1	RSV-Ld	~ 16	12 months - 23 months	x	x
	Placebo-Ld	~ 16	12 months - 23 months	x	x
2	RSV-Md	~ 16	12 months - 23 months	x	x
	Placebo-Md	~ 16	12 months - 23 months	x	x
3	RSV-Hd	~ 16	12 months - 23 months	x	x
	Placebo-Hd	~ 16	12 months - 23 months	x	x

RSV-Ld: low dose ChAd155-RSV vaccine (5 x 10^9 vp); **RSV-Md**: middle dose ChAd155-RSV vaccine (1.5 x 10^{10} vp); **RSV-Hd**: high dose ChAd155-RSV vaccine (5 x 10^{10} vp).

***The target will be to enroll up to 96 RSV-seropositive infants aged 12 to 23 months at the time of first vaccination (unless the dose escalation IDMC decision can be made based on the evaluation of accumulating safety data on fewer subjects) to ensure that at least 64 infants receive two doses of study vaccine (ChAd155-RSV or Placebo) (see Section 5.1). Then the number of subjects at each step will be at least 16 (approximately 8 subjects per group) at steps 1 and 2 and 32 (approximately 16 subjects per group at step 3)* (Amended 10 December 2017).**

Table 2 Study groups and treatment foreseen in the study

Treatment name	Volume administered	Vaccine/Product name	Study Groups					
			Step 1		Step 2		Step 3	
			RSV-Ld	Placebo-Ld	RSV-Md	Placebo-Md	RSV-Hd	Placebo-Hd
ChAd155 $5x10^9$ vp	0.5 ml	ChAd155-RSV	X					
ChAd155 $1.5x10^{10}$ vp	0.15 ml	ChAd155-RSV			X			
ChAd155 $5x10^{10}$ vp	0.5 ml	ChAd155-RSV					X	
Placebo 0.5	0.5 ml	Formulation buffer S9b		X				X
Placebo 0.15	0.15 ml	Formulation buffer S9b				X		

RSV-Ld: low dose ChAd155-RSV vaccine (5×10^9 vp); **RSV-Md**: middle dose ChAd155-RSV vaccine (1.5×10^{10} vp); **RSV-Hd**: high dose ChAd155-RSV vaccine (5×10^{10} vp).

- Control: placebo control (Formulation buffer S9b).
- Vaccination schedule(s): two IM vaccine doses administered according to a 0, 1-month schedule (i.e. at Day 0 and Day 30).
- Treatment allocation: infants will be randomized using a randomization system on internet (SBIR) at first vaccination.
- Blinding: observer-blind in Epoch 001 and single-blind in Epoch 002.

Table 3 Blinding of study epochs

Study Epochs	Blinding
Epoch 001	observer-blind
Epoch 002	single-blind

- Sampling schedule:
  - Blood samples for **biochemistry** will be taken from all infants at Screening (up to 29 days before first vaccination to Day 0) and on Day 30, and Day 60 (refer to Table 11 for the list of parameters to be tested). Blood samples for **hematology** will be taken from all infants at Screening (up to 29 days before first vaccination to Day 0) and on Day 1, Day 30, Day 31, and Day 60 (refer to Table 11 for the list of parameters to be tested). Day 7 hematology testing will be performed if any ≥ Grade 1 for platelet decrease is detected on Day 1 after vaccination, to ensure its resolution. Similarly, hematology testing on Day 37 will be performed if any ≥ Grade 1 for platelet decrease is detected on Day 31. A clinical examination with special attention given to detection of spontaneous bleeding or easy bruising and evidence of bruising or petechiae will be performed by the investigator/study staff on Day 1, Day 3 and Day 7 after each vaccination (i.e. Day 1, Day 3, Day 7, Day 31, Day 33, and Day 37). The visits on Day 3 and Day 33 may be replaced by a phone call by the investigator/study staff to inquire of the infants' general health and to solicit reports of a rash, spontaneous bleeding or easy bruising. On Day 1, and Day 31, special attention will also be given to the detection of petechiae or evidence of petechiae induced by the tourniquet. This special attention to detection of petechiae or evidence of petechiae induced by the tourniquet will also be given on Day 7 and/or Day 37 in case hematology

testing will be performed on that day. Further testing may be required to investigate a finding or guide subject management based on the investigators clinical judgment. Subjects' parent(s)/LAR(s) will be instructed to contact the investigator/study staff if their child presents symptoms of spontaneous bleeding or easy bruising or if their child develops a rash in the month following vaccination, in order to detect any thrombocytopenic petechiae or purpura. The investigator will, based on his/her medical judgement, appropriately investigate infants with clinical suspicion of low platelets.

- Refer to Section 6.6.11.2 and Figure 2 for information about the re-testing of samples in case of any Grade 1 abnormality with potential clinical relevance or any ≥ Grade 2 abnormality.
- Blood sample for **RSV serostatus** will be taken from all infants at Screening.
- Blood samples for **CMI** are limited to those in investigational sites with a laboratory in proximity capable to perform WBS necessary for the CMI assay. In those sites, samples will be taken from all subjects at Screening and on Day 30, Day 60, and Day 365.

  Note: Blood samples for CMI taken on Day 7 and Day 37 for subjects enrolled under protocol amendment 1 will still be assessed for CMI.
- Blood samples for **humoral immunogenicity** will be taken from all subjects at Screening and on Day 30, Day 60, and Day 365.
- Blood sample for **assessment of mechanism of illness (potential ERD)** will be taken from subjects hospitalized for LRTI (only for RSV-positive subjects using a locally available RSV test).
- Nasal swab: there will be monthly nasal swab to detect asymptomatic RSV infections during the RSV season and if following active or a passive surveillance contacts, a subject presents symptoms of RTI, a nasal swab will be collected.

- Study visits: Visit 1 (Day 0), Visit 5 (Day 30), Visit 9 (Day 60) and Visit 10 (Day 365) must be performed at the investigators clinical facility. Visit 2 (Day 1), Visit 4 (Day 7), Visit 6 (Day 31), Visit 8 (Day 37) and Visit 11 (Day 730) (no blood sampling for immune response and no vaccine administration) may take place in the subject's home or at the investigators clinical facility as appropriate to the circumstances in the judgment of the investigator. Visit 3 (Day 3) and Visit 7 (Day 33) may take place at the investigators clinical facility or the investigator/clinical staff may call the subjects' parent(s)/LAR(s), as appropriate to the circumstances in the judgment of the investigator.
- Type of study: e.g. self-contained
- Data collection: Electronic Case Report Form (eCRF).
- Safety monitoring: iSRC and IDMC (refer to Section 9.10).
- Surveillance for RSV-RTI, difficulty in breathing and wheezing episodes.

Surveillance period will be carried out from Visit 1 (after Dose 1) until Visit 11 (Day 730). In order to detect asymptomatic RSV-RTI, monthly nasal swabs for analysis at sponsor laboratory will be performed for all subjects during the RSV season. In order to timely detect RSV-RTI and to ensure cases are timely captured by the study sites, both active and passive surveillance will be conducted:

- **Passive surveillance**: Parent(s)/LAR(s) are instructed to contact the investigator/study staff as soon as the subject experiences new RTI symptoms (cough, runny nose or blocked nose) or worsening of RTI symptoms, or in case of difficulty in breathing or wheezing.
- **Active surveillance**: parent(s)/LAR(s) of all the subjects will be contacted by the investigator/study staff on a regular basis (weekly during the RSV season and every month outside the RSV season) to identify any potential RSV-RTI and to remind the parent(s)/LAR(s) of the subjects to report any new occurrence of RTI symptoms (cough, runny nose, blocked nose) or worsening of RTI symptoms, or in case of difficulty in breathing or wheezing as soon as possible.

Refer to Section 9.2 for more information about active and passive surveillance.

4. CASE DEFINITION OF RESPIRATORY TRACT INFECTION ASSOCIATED WITH RSV INFECTION

It should be noted that for the reporting of safety (unsolicited AEs, AEs of specific interest and serious adverse events [SAEs]), the investigator will use all available information and his/her medical judgment to make diagnoses. The case definitions presented below will be used for a safety analysis complementary to standard safety tabulations of events according to Medical Dictionary for Regulatory Activities (MedDRA) preferred terms.

4.1. RTI case definitions

During the analysis of the study, all cases identified during the surveillance of RSV-RTI will be definitively classified as either RTI, LRTI, severe LRTI or very severe LRTI according to the standardized case definitions (see Table 4) based on the available World Health Organization (WHO) case definitions.

Table 4 Case definitions for data analysis

RSV-RTI	Runny nose OR blocked nose OR cough AND Confirmed RSV infection [4]
RSV-LRTI	History of cough OR difficulty breathing [1] AND SpO_2 < 95% [2], OR RR increase [3] AND Confirmed RSV infection [4]
RSV-severe LRTI	Meeting the case definition of RSV-LRTI AND SpO_2 < 93% [2], OR lower chest wall in-drawing
RSV-very severe LRTI	Meeting the case definition of RSV-LRTI AND SpO_2 < 90% [2], OR inability to feed, OR failure to respond / unconscious
RSV hospitalization	Confirmed RSV infection [5] AND Hospitalized for acute medical condition [6]
All-cause LRTI	History of cough OR difficulty breathing [1] AND SpO_2 < 95% [2], OR RR increase [3]

Definitions based on [Modjarrad, 2016]
LRTI = lower respiratory tract infections; RR = respiratory rate; RTI = respiratory tract infections; SpO_2 = blood oxygen saturation.
[1] Based on history reported by parents/LARs and includes difficulty breathing (e.g. showing signs of wheezing or stridor, tachypnoea, flaring [of nostrils], chest in-drawing, apnoea) associated with nasal obstruction.
[2] For blood oxygen saturation (SpO_2), the lowest value monitored will be used.
[3] RR increase defined as $\geq$ 40/minute (12 months of age or above).
[4] RSV infection confirmed on nasal swab positive for RSV A or B by quantitative Reverse Transcription Polymerase Chain Reaction (qRT-PCR).
[5] RSV sampling and testing is based on medical judgment of medical practitioner or driven by algorithm.
[6] Hospitalization is defined as a medical decision that the infant requires admission for observation or treatment.

4.2. RTI episode

4.2.1. Start date of the RTI episode

The start date of the RTI episode is defined as the point at which the first symptoms of cough, runny nose, blocked nose, wheezing, or difficulty breathing were observed.

4.2.2. End date of the RTI episode

The end date of the RTI episode is defined as the point at which the child is considered symptom-free of cough, runny nose, blocked nose, wheezing, or difficulty breathing.

5. STUDY COHORT

5.1. Number of subjects

The target will be to enroll ***up to*** 96 RSV-seropositive infants aged 12 to 23 months at the time of first vaccination ***(unless the dose escalation IDMC decision can be made based on the evaluation of accumulating safety data on fewer subjects*** to ensure that ***at least 64*** infants receive ***two*** dose***s*** of study vaccine ***(ChAd155-RSV or Placebo)*** **(Amended 10 December 2017)**. Refer to Section 11.4 for the determination of sample size.

Overview of the screening plan:

Upon completion of all screening procedures (Refer to Table 5), the investigator will review the inclusion/exclusion criteria for each infant. Infants meeting all eligibility criteria will be enrolled in the study. Their screening information will be recorded on the appropriate screen of the eCRF.

If the investigator believes there is a reasonable reason to do so, screening procedures may be repeated once only. All screening procedures will be repeated except the blood sampling for CMI response and humoral response (neutralizing antibody titers against RSV-A, RSV F protein antibody concentrations and palivizumab-competing antibody concentrations). Blood sampling to assess RSV serostatus (IBL International kit) might not be repeated if the infant has been confirmed RSV-seropositive during the first screening.

5.2. Inclusion criteria for enrolment

Deviations from inclusion criteria are not allowed because they can potentially jeopardize the scientific integrity of the study, regulatory acceptability or subject safety. Therefore, adherence to the criteria as specified in the protocol is essential.

All subjects must satisfy ALL the following criteria at study entry:

- Subjects' parent(s)/LAR(s) who, in the opinion of the investigator, can and will comply with the requirements of the protocol (e.g. completion of the diary cards, return for follow-up visits).
- Written informed consent obtained from the parent(s)/LAR(s) of the subject prior to performance of any study specific procedure.
- A male or female between, and including, 12 and 23 months (from the day the infant becomes 12 months of age until the day before the infant achieves 24 months of age) at the time of the first vaccination.
- Healthy subjects as established by medical history and clinical examination before entering into the study.
- Seropositive for RSV as determined by IBL International kit.
- Born full-term (i.e. after a gestation period of 37 to less than 42 completed weeks) with a minimum birth weight of 2.5 kg. (Required for Spain)
- Subjects' parent(s)/LAR(s) need to have access to a consistent mean of telephone contact (e.g. land line or mobile) or computer.

5.3. Exclusion criteria for enrolment

Deviations from exclusion criteria are not allowed because they can potentially jeopardize the scientific integrity of the study, regulatory acceptability or subject safety. Therefore, adherence to the criteria as specified in the protocol is essential.

The following criteria should be checked at the time of study entry. If ANY exclusion criterion applies, the subject must not be included in the study:

- Child in care.

  Please refer to the glossary of terms for the definition of child in care.
- Use of any investigational or non-registered product (drug or vaccine) other than the study vaccine during the period starting 30 days before the first dose of study vaccine (Day -29 to Day 0), or planned use during the study period.
- Any medical condition that in the judgment of the investigator would make IM injection unsafe.
- Chronic administration (defined as more than 14 days in total) of immunosuppressants or other immune-modifying drugs during the period starting six months prior to the first vaccine. For corticosteroids, this will mean prednisone (≥ 0.5 mg/kg/day (for pediatric subjects), or equivalent. Inhaled and topical steroids are allowed.
- Administration of long-acting immune-modifying drugs (e.g. infliximab) or planned administration at any time during the study period.

- Administration of immunoglobulins and/or any blood products during the period starting three months before the first dose of study vaccine or planned administration during the study period.
- Planned administration/administration of a vaccine not foreseen by the study protocol in the period starting 30 days before the first dose and ending 30 days after the last dose of vaccine administration, with the exception of scheduled routine pediatric vaccines which may be administered ≥ 14 days before a dose or ≥ 7 days after a dose.
- Acute or chronic, clinically significant pulmonary, cardiovascular, hepatic or renal functional abnormality, as determined by physical examination or laboratory screening tests.
- Serious chronic illness.
- Major congenital defects.
- History of any neurological disorders or seizures.
- History of or current autoimmune disease.
- History of recurrent wheezing. (Wheezing should have been verified on auscultation by doctor).
- History of chronic cough (4 weeks or more duration).
- Previous hospitalization for respiratory illnesses.
- History of thrombocytopenia.
- History of anemia.
- Previous, current or planned administration of *Synagis* (palivizumab).
- Neurological complications following any prior vaccination.
- Born to a mother known or suspected to be HIV-positive (no laboratory testing required).
- Any confirmed or suspected immunosuppressive or immunodeficient condition, based on medical history and physical examination (no laboratory testing required).
- Family history of congenital or hereditary immunodeficiency.
- Previous vaccination with a recombinant simian or human adenoviral vaccine.
- History of any reaction or hypersensitivity likely to be exacerbated by any component of the vaccine.
- Hypersensitivity to latex.
- Current severe eczema.
- Acute disease and/or fever at the time of enrolment.
  - Fever is defined as temperature ≥ 37.5°C/99.5°F for oral, axillary or tympanic route, or ≥ 38.0°C/100.4°F for rectal route. The preferred route for recording temperature in this study will be axillary.

  - Clinically significant upper respiratory tract infection.
  - Subjects with a minor illness (such as mild diarrhea) without fever may, be enrolled at the discretion of the investigator.

- Any clinically significant Grade 1* or any ≥ Grade 2* hematological or biochemical laboratory abnormality detected at the last screening blood sampling.

  * Refer to Appendix C. For Grade 1 laboratory abnormalities, the investigator should use his/her clinical judgment to decide which ones are clinically relevant. Infants with hematological/biochemical values out of normal range which are expected to be temporary, may be re-screened at a later date.

- Any other conditions that the investigator judges may interfere with study procedures (e.g. drawing blood) or findings (e.g. immune response).
- Any conditions that could constitute a risk for the subjects while participating to this study.
- Weight below the fifth percentile of the local weight-for-age curve.
- Concurrently participating in another clinical study, at any time during the study period, in which the subject has been or will be exposed to an investigational or a non-investigational vaccine/product (pharmaceutical product or device).
- Planned move to a location that will prohibit participating in the trial until study end.

6. CONDUCT OF THE STUDY

6.1. Regulatory and ethical considerations, including the informed consent process

The study will be conducted in accordance with all applicable regulatory requirements.

The study will be conducted in accordance with the ICH Guideline for Good Clinical Practice (GCP), all applicable subject privacy requirements and the guiding principles of the Declaration of Helsinki.

The study has been designed and will be conducted in accordance with the ICH Harmonized Tripartite Guideline for clinical investigation of medicinal products in the pediatric population (ICH E11) and all other applicable ethical guidelines.

GSK will obtain favorable opinion/approval to conduct the study from the appropriate regulatory agency, in accordance with applicable regulatory requirements, prior to a site initiating the study in that country.

Conduct of the study includes, but is not limited to, the following:

- Institutional Review Board (IRB)/Independent Ethics Committee (IEC) review and favorable opinion/approval of study protocol and any subsequent amendments.
- Subject's parent(s)/LAR(s) informed consent.
- Investigator reporting requirements as stated in the protocol.

GSK will provide full details of the above procedures to the investigator, either verbally, in writing, or both.

Freely given and written or witnessed/ thumb printed informed consent must be obtained from each subject's parent(s)/LAR(s) prior to participation in the study.

GSK Biologicals will prepare a model Informed Consent Form (ICF) which will embody the ICH GCP and GSK Biologicals required elements. While it is strongly recommended that this model ICF is to be followed as closely as possible, the informed consent requirements given in this document are not intended to pre-empt any local regulations which require additional information to be disclosed for informed consent to be legally effective. Clinical judgment, local regulations and requirements should guide the final structure and content of the local version of the ICF.

The investigator has the final responsibility for the final presentation of the ICF, respecting the mandatory requirements of local regulations. The ICF generated by the investigator with the assistance of the sponsor's representative must be acceptable to GSK Biologicals and be approved (along with the protocol, and any other necessary documentation) by the IRB/IEC.

6.2. Subject identification and randomization of treatment

6.2.1. Subject identification

Subject identification numbers will be assigned sequentially to the subjects who have consented to participate in the study, according to the range of subject identification numbers allocated to each study center.

6.2.2. Randomization of treatment

6.2.2.1. Randomization of supplies

The randomization of supplies within blocks will be performed at GSK Biologicals, using MATerial EXcellence (MATEX), a program developed for use in Statistical Analysis System (SAS®) (Cary, NC, USA) by GSK Biologicals. Entire blocks will be shipped to the study centers /warehouse(s).

6.2.2.2. Treatment allocation to the subject

The treatment numbers will be allocated by dose.

6.2.2.2.1. Study group and treatment number allocation

The target will be to enroll ***up to*** 96 RSV-seropositive infants aged 12 to 23 months at the time of first vaccination ***(unless the dose escalation IDMC decision can be made based on the evaluation of accumulating safety data on fewer subjects*** to ensure that ***at least 64*** infants receive ***two*** dose***s*** of study vaccine ***(ChAd155-RSV or Placebo) (see Section 5.1)*** **(Amended 10 December 2017)**. Infants will be randomly assigned to two study groups per step in a (1:1) ratio (approximately 16 subjects in each group).

- In **Step 1**, ***between*** approximately ***8 and*** 16 RSV-seropositive infants will be randomly assigned to receive low dose ChAd155-RSV vaccine (5×10^9 vp [RSV-Ld]; 0.5 mL) and ***between*** approximately ***8 and*** 16 RSV-seropositive infants will be randomly assigned to receive placebo (0.5 mL). **(Amended 10 December 2017)**
- In **Step 2**, ***between*** approximately ***8 and*** 16 RSV-seropositive infants will be randomly assigned to receive middle dose ChAd155-RSV vaccine (1.5×10^{10} vp [RSV-Md]; 0.15 mL) and ***between*** approximately ***8 and*** 16 RSV-seropositive infants will be randomly assigned to receive placebo (0.15 mL). **(Amended 10 December 2017)**
- In **Step 3,** approximately 16 RSV-seropositive infants will be randomly assigned to receive high dose ChAd155-RSV vaccine (5×10^{10} vp [RSV-Hd]; 0.5 mL) and approximately 16 RSV-seropositive infants will be randomly assigned to receive placebo (0.5 mL).

Allocation of the subject to a study group at the investigator site will be performed using a randomization system on internet (SBIR). Within each step, the randomization algorithm will use a minimization procedure accounting for center.

After obtaining the signed and dated ICF from the subject's parent(s)/LAR(s) and having checked the eligibility of the subject, the study staff in charge of the vaccine administration will access SBIR. Upon providing the subject identification number, the randomization system will determine the study group and will provide the treatment number to be used for the first dose.

The number of each administered treatment must be recorded in the eCRF on the Vaccine Administration screen.

When SBIR is not available, please refer to the SBIR user guide or the SPM for specific instructions.

6.2.2.2.2. Treatment number allocation for subsequent doses

For each dose subsequent to the first dose, the study staff in charge of the vaccine administration will access SBIR, provide the subject identification number, and the system will provide a treatment number consistent with the allocated study group.

The number of each administered treatment must be recorded in the eCRF on the Vaccine Administration screen.

6.3. Method of blinding

Data will be collected in an observer-blind manner. By observer-blind, it is meant that during the course of the study, the vaccine recipient and those responsible for the evaluation of any study endpoint (e.g. safety, reactogenicity) will all be unaware of which vaccine was administered. To do so, vaccine preparation and administration will be done by authorized medical personnel who will not participate in any of the study clinical evaluation assays.

When all data up to Day 60 are available, a statistical analysis will be performed. This analysis may lead to the unblinding of some subjects. As a consequence, after Day 60, the study cannot be considered as observer-blind, but will be conducted in a single-blind manner, with subjects' parent(s)/ LAR(s) remaining blinded up to study end (Day 730). The investigators will not have access to the treatment allocation up to study end (Day 730).

The laboratory in charge of the laboratory testing will be blinded to the treatment, and codes will be used to link the subject and study (without any link to the treatment attributed to the subject) to each sample.

6.4. General study aspects

Supplementary study conduct information not mandated to be present in this protocol is provided in the accompanying SPM. The SPM provides the investigator and the site personnel with administrative and detailed technical information that does not impact the safety of the subjects.

Vaccination will be limited to a maximum of 10 infants per day. In the same investigational center, infants will be vaccinated sequentially, separated by a minimum interval of 60 minutes. In addition, to monitor delayed potential reactions following vaccination, there will be a 48-hour observation period following Dose 1 and Dose 2 administration to the first eight infants (corresponding to three to five ChAd155-RSV vaccine recipients) in each Step (1, 2, and 3) before continuing vaccination of more infants (i.e. from the ninth infant). From the ninth infant, infants will be vaccinated sequentially, separated by a minimum interval of 60 minutes.

Refer to Section 9.10 for information about limited vaccination, holding rules, safety monitoring and safety evaluation by iSRC and IDMC.

6.5. Outline of study procedures

Table 5 List of study procedures

Epoch	Epoch 001										Epoch 002					
Age	12-23 mth										24-35 mth	36-47 mth				
Type of contact	Screening	Visit 1	Visit 2#	Visit 3##	Visit 4#	Visit 5	Visit 6#	Visit 7##	Visit 8#	Visit 9	Visit 10	Visit 11#	Unscheduled visit for safety*	Contact for active/passive surveillance**	Surveillance for asymptomatic RSV-RTI***	Assessment of potential RSV-RTI****
Timepoints	D-29 to D0	D0	D1	D3	D7	D30	D31	D33	D37	D60	D365	D730		Monthly or Weekly	Monthly	
Informed consent	●															
Check inclusion/exclusion criteria	●	O														
Collect demographic data	●[a]															
Collect birth weight and gestation at birth in weeks	●															
Medical history	●															
Physical examination[b]	●	O	O		O	O	O		O	O	O	O	O		O	●
Growth monitoring[c]	●					●				●	●	●				
Check contraindications and warnings and precautions		O				O										
Pre-vaccination body temperature		●				●										
Randomization		O														
Vaccine administration		●				●										
Recording of administered treatment number		●				●										
60 minutes post-vaccination observation[d]		●				●										
Blood sampling for RSV serostatus (1.0 mL)	●															

Epoch	Epoch 001										Epoch 002					
Age	12-23 mth										24-35 mth	36-47 mth				
Type of contact	Screening	Visit 1	Visit 2#	Visit 3##	Visit 4#	Visit 5	Visit 6#	Visit 7##	Visit 8#	Visit 9	Visit 10	Visit 11#	Unscheduled visit for safety*	Contact for active/passive surveillance**	Surveillance for asymptomatic RSV-RTI***	Assessment of potential RSV-RTI****
Timepoints	D-29 to D0	D0	D1	D3	D7	D30	D31	D33	D37	D60	D365	D730		Monthly or Weekly	Monthly	
Blood sampling for assessment of mechanisms of illness (potential ERD; 2.5 mL)																●[l]
Blood sampling for hematology (1.2 mL)	●[e]		●[f]		●[f,p]	●[f,g]	●[f]		●[f,p]	●[f]			●[f]			
Blood sampling for biochemistry (1.1 mL)	●[e]					●[f,g]				●[f]			●[f]			
Blood sampling for CMI response (2.0 mL) [o]	●[r]					●[g]				●	●					
Blood sampling for humoral response (2.5 mL)	●[r]					●[g]				●	●					
Detection of spontaneous bleeding or easy bruising and evidence of bruising or petechiae [q]			●	●	●		●	●	●							
Examination of limb for petechiae induced by the tourniquet			●		●		●		●							
Surveillance for RSV-RTI, difficulty in breathing and wheezing		O	O	O	O	O	O	O	O	O	O	O		●	●	
Documentation of symptoms and signs of RTI[j]																●
Nasal swab for central testings															●[m]	●[h]
Specimen for local testings																●[n]
Distribution of RTI episode cards[k]		O	O		O	O	O		O	O	O					
Collection of completed RTI episode cards[k]			O		O	O	O		O	O	O	O	O		O	O

Epoch	Epoch 001										Epoch 002					
Age	12-23 mth										24-35 mth	36-47 mth				
Type of contact	Screening	Visit 1	Visit 2#	Visit 3##	Visit 4#	Visit 5	Visit 6#	Visit 7##	Visit 8#	Visit 9	Visit 10	Visit 11#	Unscheduled visit for safety*	Contact for active/passive surveillance**	Surveillance for asymptomatic RSV-RTI***	Assessment of potential RSV-RTI****
Timepoints	D-29 to D0	D0	D1	D3	D7	D30	D31	D33	D37	D60	D365	D730		Monthly or Weekly	Monthly	
Transcription of completed RTI episode cards			●		●	●	●		●	●	●	●	O		●	●
Record any concomitant medications/vaccinations		●	●		●	●	●		●	●	●	●	●		●	●
Distribution of the subject card		O														
Distribution of diary card		O				O										
Return of diary card					O	O			O	O						
Diary card transcription by investigator					●	●			●	●						
Recording of solicited AEs (Day 0-6)		●	●		●	●	●		●				●[i]			
Recording of unsolicited AEs (Day 0-29)		●	●	●	●	●	●	●	●	●			●[i]			
Recording of AE leading to study withdrawal		●	●	●	●	●	●	●	●	●	●	●	●	●	●	●
Recording of AE of specific interest (RSV-LRTI)		●	●	●	●	●	●	●	●	●	●	●	●	●	●	●
Recording of AE of specific interest (spontaneous or excessive bleeding)		●	●	●	●	●	●	●	●	●			●	●	●	●
Recording of SAEs		●	●	●	●	●	●	●	●	●	●	●	●	●	●	●
Recording of SAEs related to study participation, or to a concurrent GSK medication/vaccine	●	●	●	●	●	●	●	●	●	●	●	●	●	●	●	●
Screening conclusion	●															
Study conclusion												●				
Signing of investigator signature form by investigator after Screening and before each analysis	●									●	●	●				

● is used to indicate a study procedure that requires documentation in the individual eCRF.

O is used to indicate a study procedure that does not require documentation in the individual eCRF.
Note: the double-bordered lines following Visit 9 (Day 60), Visit 10 (Day 365) and Visit 11 (Day 730) indicate the statistical analyses which will be performed. After Visit 9 (Day 60) the study will be conducted in a single-blind manner, with patients remaining blinded up to study end (Day 730). The investigators will not have access to the treatment allocation up to study end (Day 730).
* This visit is applicable for infants with hematological/biochemical values out of normal range or for further evaluation of clinical suspicion of low platelet count.
** Active contacts for surveillance of RSV-RTI will take place weekly during the RSV season and every month outside the RSV season (refer to Section 9.2). Passive phone contacts from the subjects' parent(s)/LAR(s) to the investigator will take place when symptoms occur.
*** In order to detect asymptomatic RSV infection, monthly visits will be performed during the RSV season.
**** This visit is only applicable for infants with potential RSV-RTI.
[#] Visit 2 (Day 1), Visit 4 (Day 7), Visit 6 (Day 31), Visit 8 (Day 37) and Visit 11 (Day 730) may take place in the subject's home or at the investigators clinical facility as appropriate to the circumstances in the judgment of the investigator.
[##] On Day 3 and Day 33, a visit may take place at the investigators clinical facility or the investigator /clinical staff may call the subjects' parent(s)/LAR(s), as appropriate to the circumstances in the judgment of the investigator.
[a] Recording of demographic data includes date of birth, gender, geographic ancestry, and ethnicity.
[b] At Screening, perform a complete physical examination, including assessment of vital signs (body temperature, heart rate [HR], and RR). At subsequent study visits, perform a physical examination only if the subject's parent(s)/LAR(s) indicate(s) during questioning that there might be some underlying pathology(ies) or if deemed necessary.
[c] Growth monitoring includes weight and length (for infants < 24 months of age) or height (for subjects ≥ 24 months of age).
[d] Infants will need to remain under observation (visual follow-up as well as measurement of resting vital signs) at the study site for at least 60 minutes after vaccination. Vital signs are body temperature, HR and RR. Vital signs are measured preferably when the infant is calm.
[e] At Screening, for infants with hematological/biochemical values out of normal range which are expected to be temporary, a re-screening visit may be scheduled during which blood sample collection for hematology/biochemistry will be repeated (maximum one re-screening visit per infant is allowed; blood for CMI response and humoral response will not be re-sampled at the re-screening visit, if already done before).
[f] If any Grade 1 abnormality with potential clinical relevance (according to investigators judgment) or any ≥ Grade 2 abnormality is detected, or for further evaluation of clinical suspicion of low platelet count, refer to Section 6.6.11.2 and Figure 2 for re-test.
[g] Blood samples to be taken before vaccination.
[h] If during passive or active surveillance contact, the investigator/study staff assesses that an infant presents a potential RSV-RTI (refer to Table 4 for definition), the potential RSV infection will be assessed by quantitative RT-PCR.
[i] Only when the unscheduled visit occurs within the AE reporting time frame, i.e. from Day 0 to Day 6 for solicited AEs and from Day 0 to Day 29 for unsolicited AEs.
[j] Signs and symptoms to be recorded in the eCRF are listed in the SPM.
[k] Subject's parent(s)/LAR(s) will be instructed to record on the RTI episode card the start date and the end date of the following symptoms (cough, runny nose, blocked nose, difficulty in breathing, or wheezing) and to return it to the investigator at the next visit or by mail (e-mail or postal mail).
[l] Blood sample collected for subjects hospitalized for LRTI(only for RSV-positive subjects using a locally available RSV test); Refer to Section 6.7.3.
[m] Nasal swab collected at the monthly surveillance for asymptomatic RSV-RTI will be tested by quantitative RT-PCR.
[n] For RSV testing and respiratory viral panel (RVP) at a local routine laboratory, where available.
[o] In investigational sites with a laboratory in proximity capable to perform WBS necessary for the CMI assay.
[p] Day 7 hematology testing will be performed if any ≥ Grade 1 for platelet decrease is detected on Day 1 after vaccination. Similarly, hematology testing on Day 37 will be performed if any ≥ Grade 1 for platelet decrease is detected on Day 31.

[q] A clinical examination with special attention given to detection of spontaneous bleeding or easy bruising and evidence of bruising or petechiae will be performed by the investigator/study staff on Day 1, Day 3 and Day 7 after each vaccination (i.e. Day 1, Day 3, Day 7, Day 31, Day 33 and Day 37). The visits on Day 3 and Day 33 may be replaced by a phone call by the investigator/study staff to inquire of the infants' general health and to solicit reports of a rash, spontaneous bleeding or easy bruising. Subjects' parent(s)/LAR(s) will be instructed to contact the investigator/study staff if their child presents symptoms of spontaneous bleeding or easy bruising or if their child develops a rash in the month following vaccination, in order to detect any thrombocytopenic petechiae or purpura.

[r] Blood samples for CMI and for humoral immunogenicity may be postponed until after the RSV serostatus is known and eligibility is confirmed up to or on Day 0 (but before vaccination).

Table 6 Intervals between study visits

Interval	Optimal length of interval	Allowed interval
Screening → Visit 1 (Day 0)	1 - 30 days[1]	-
Visit 1 (Day 0) → Visit 2 (Day 1)	1 day	-
Visit 1 (Day 0) → Visit 3 (Day 3)	3 days	3 - 4 days
Visit 1 (Day 0) → Visit 4 (Day 7)	7 days	7 - 10 days
Visit 1 (Day 0) → Visit 5 (Day 30)	30 days	23 - 36 days[2]
Visit 5 (Day 30) → Visit 6 (Day 31)	1 day	-
Visit 5 (Day 30) → Visit 7 (Day 33)	3 days	3 - 4 days
Visit 5 (Day 30) → Visit 8 (Day 37)	7 days	7 - 10 days
Visit 5 (Day 30) → Visit 9 (Day 60)	30 days	28 - 35 days[2]
Visit 1 (Day 0) → Visit 10 (Day 365)	365 days	335 - 395 days[2]
Visit 1 (Day 0) → Visit 11 (Day 730)	730 days	700 - 760 days

[1] Visit 1 should take place no longer than 30 days after the Screening visit. When applicable, a re-screening visit may be scheduled at any time (but only once to assess eligibility; blood for CMI response and humoral response will not be re-sampled, if already done before). All screening procedures need to be performed within 30 days of Visit 1.
[2] Immunogenicity data from blood samples collected outside this interval will not be eligible for inclusion in the according-to-protocol (ATP) cohort for analysis of immunogenicity.

6.6. Detailed description of study procedures

6.6.1. Informed consent

The signed/witnessed/thumb printed informed consent of the subject's parent(s)/LAR(s) must be obtained before study participation. Refer to Section 6.1 for the requirements on how to obtain informed consent.

6.6.2. Check inclusion and exclusion criteria

Check all inclusion and exclusion criteria as described in Sections 5.2 and 5.3 before enrolment.

6.6.3. Collect demographic data

Record demographic data such as date of birth (day, month and year), gender, geographic ancestry, and ethnicity in the subject's eCRF. Collect and record the birth weight and gestation at birth (in weeks) in the eCRF.

6.6.4. Medical history

Obtain the subject's medical history by interview and/or review of the subject's medical records and record any pre-existing conditions or signs and/or symptoms present in a subject prior to the first study vaccination in the eCRF.

6.6.5. Physical examination

At Screening, perform a physical examination of the subject, including assessment of vital signs preferably when the infant is calm. Vital signs are body temperature, heart rate (HR) and RR. Collected information needs to be recorded in the eCRF.

At subsequent study visits, perform a physical examination only if the subject's parent(s)/LAR(s) indicate(s) during questioning that there might be some underlying pathology(ies) or if deemed necessary.

Treatment of any abnormality observed during physical examination has to be performed according to local medical practice outside this study or by referral to an appropriate health care provider.

6.6.6. Growth monitoring

Growth monitoring includes weight and length (for infants < 24 months of age) or height (for subjects ≥ 24 months of age; [United Nations, 1986]). Collected information needs to be recorded in the eCRF.

6.6.7. Check contraindications, warnings and precautions to vaccination

Contraindications, warnings and precautions to vaccination must be checked at the beginning of each vaccination visit. Refer to Section 7.5 for more details.

6.6.8. Assess pre-vaccination body temperature

The axillary, rectal, oral, or tympanic body temperature of all subjects needs to be measured prior to any study vaccine administration **(Amended 10 December 2017)**. The preferred route for recording temperature in this study will be axillary. If the subject has fever (fever is defined as temperature ≥ 37.5°C/99.5°F for oral, axillary or tympanic route, or ≥ 38.0°C/100.4°F for rectal route) on the day of vaccination, the vaccination visit will be rescheduled within the allowed interval for this visit (see Table 6).

6.6.9. Randomization

Study group and treatment number allocation will be performed as described in Section 6.2.2. The number of each administered treatment must be recorded in the eCRF.

6.6.10. Study Vaccines administration

- Across each steps, vaccination will be limited to a maximum of 10 infants per day. In the same investigational center, infants will be vaccinated sequentially, separated by a minimum interval of 60 minutes. In addition, to monitor delayed potential reactions following vaccination, there will be a 48-hour observation period following Dose 1 and Dose 2 administration to the first eight infants (corresponding to three to five

ChAd155-RSV vaccine recipients) in each Step (1, 2, and 3) before continuing vaccination of more infants (Refer to Section 9.10.1).

- After completing all prerequisite procedures prior to vaccination, one dose of study vaccine will be administered via IM injection in the deltoid (if the muscle size is adequate; otherwise injections might be done in the anterolateral thigh). The site of administration of Dose 2 should preferably remain the same as the site of administration of Dose 1. Refer to Section 7.3 for detailed description of the vaccines administration procedure. If the investigator or delegate determines that the infant's health on the day of administration temporarily precludes vaccine administration, the visit will be rescheduled within the allowed interval for this visit (refer to Table 6).
- The subjects will be observed closely (visual follow-up as well as measurement of resting vital signs) for at least 60 minutes following the administration of the vaccines, with appropriate medical treatment readily available in case of anaphylaxis. Vital signs are body temperature, HR and RR. Vital signs are preferably measured when the infant is calm.

6.6.11. Sampling

Refer to the Module on Biospecimen Management in the SPM for detailed instructions for the collection, handling and processing of the samples.

To minimize any development of needle aversion, pain relief by means of a topical local anesthetic may be offered to infants prior to any blood sampling requested by the protocol at the discretion of the investigator.

6.6.11.1. Blood sampling for RSV serostatus, humoral immunity, and cell-mediated immunity

Blood samples will be taken during certain study visits as specified in Section 6.5 List of Study Procedures.

- A volume of maximum 1.0 mL of whole blood (to provide at least 300 µL of serum) should be drawn from all infants for analysis of their RSV serostatus at the screening visit. After centrifugation, serum samples should be kept at –20°C/ –4°F or below. RSV serostatus assessment will be performed in a laboratory designated by GSK Biologicals using an ELISA commercial kit (refer to Table 9). Refer to the SPM for more details on sample storage conditions.
- A volume of 2.0 mL of whole blood should be drawn from all infants for analysis of CMI response at each pre-defined timepoint, in investigational sites with a laboratory in proximity capable to perform WBS necessary for the CMI assay. Refer to the SPM for more details on sample storage conditions.
- A volume of 2.5 mL of whole blood (to provide at least 800 µL of serum) should be drawn from all infants for analysis of humoral response at each pre-defined timepoint. After centrifugation, serum samples should be kept at –20°C/ –4°F or below until shipment. Refer to the SPM for more details on sample storage conditions.

- Blood sampling for CMI and humoral response will not be repeated in case of insufficient volume, at any timepoint.

6.6.11.2. Blood sampling for hematology and biochemistry

- A volume of maximum 1.2 mL of whole blood should be drawn from all infants for analysis of the hematology parameters (refer to Table 11) at each pre-defined timepoint.
- A volume of maximum 1.1 mL of whole blood should be drawn from all infants for analysis of the biochemistry parameters (refer to Table 11) at each pre-defined timepoint.

  Hematology and biochemistry parameters assessment will be performed according to laboratory practices.

  Day 7 hematology testing will be performed if any ≥ Grade 1 for platelet decrease is detected on Day 1. Similarly, hematology testing on Day 37 will be performed if any ≥ Grade 1 for platelet decrease is detected on Day 31 (refer to Figure 2 and Appendix C).

  If any Grade 1 abnormality with potential clinical relevance (according to investigators judgment, refer to the glossary of terms) or any ≥ Grade 2 abnormality is detected (refer to Figure 2 and Appendix C):

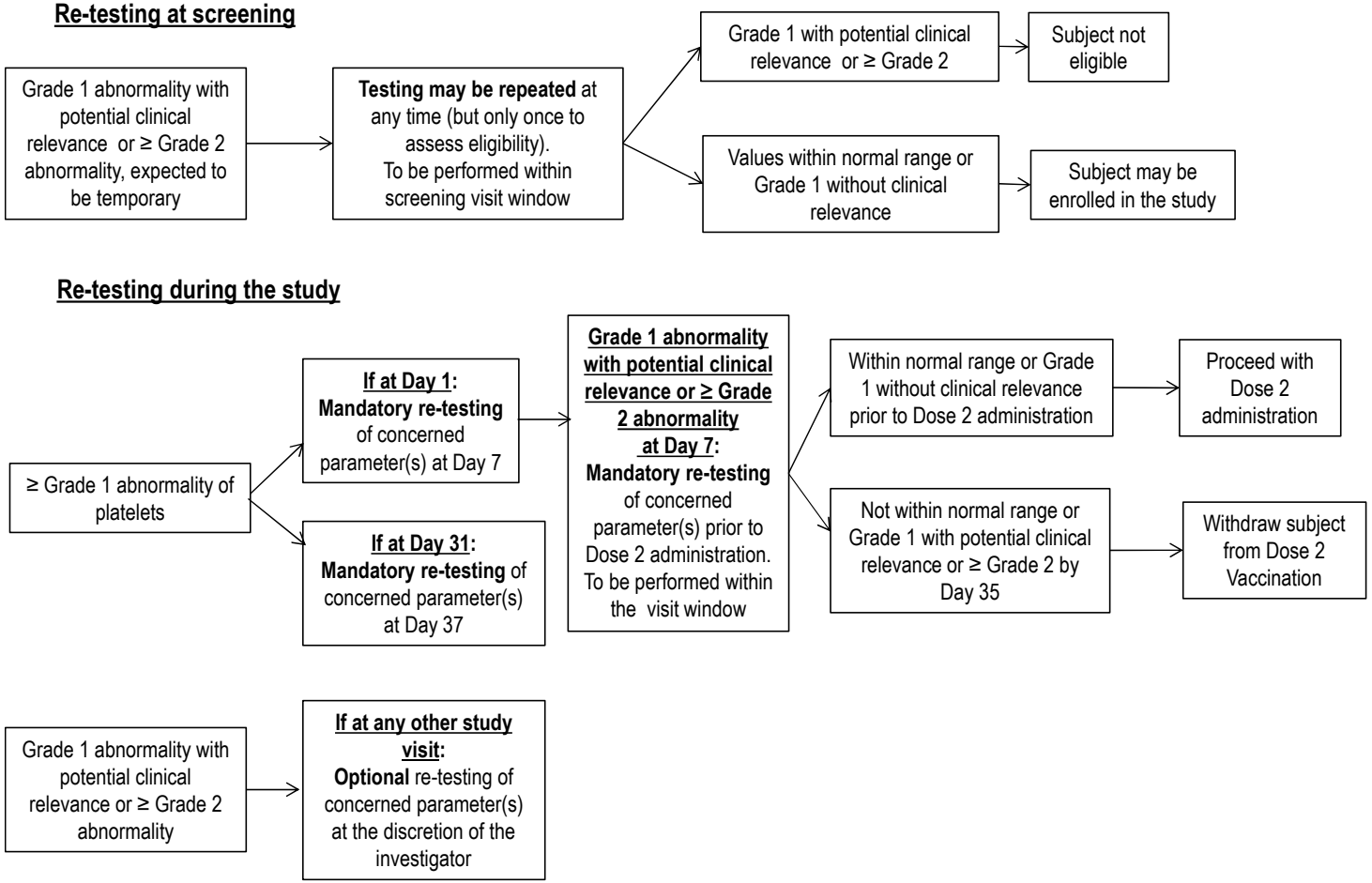
  – at **Screening**, for infants with hematological/biochemical values out of normal range which are expected to be temporary, a re-screening visit may be scheduled during which blood sample collection for hematology/biochemistry will be repeated (maximum one re-screening visit per infant is allowed within the interval planned in Table 6).

  – **at Day 7**, the relevant hematology/biochemistry parameter(s) **must** be re-tested before administration of vaccine Dose 2, either during an unscheduled visit or at the Dose 2 vaccination visit itself (provided results are obtained before vaccination). Only if the concerned parameter(s) is/are within the acceptable range (i.e. within normal range or Grade 1 without clinical relevance), vaccine Dose 2 can be administered. If the concerned parameter(s) is/are not within the acceptable range within the allowed interval for the second dosing visit, Visit 5, the infant will not receive Dose 2 but should still continue the study for safety follow-up (up to Day 730).

  – **at Days 30, 37 and 60** the relevant hematology/biochemistry parameter(s) **may** be re-tested as per the opinion of the investigator.

Note: For safety reasons and specifically for further investigation of an AE of specific interest (suspected bleeding or low platelet count), blood can be re-sampled for hematology and biochemistry assessment.

Figure 2 Schematic overview of required additional hematology and biochemistry testing during the study



Note: For safety reasons and specifically for further investigation of an AE of specific interest (suspected bleeding or low platelet count), blood can be re-sampled for hematology and biochemistry assessment.

6.6.11.3. Nasal swab and other specimen for local assay

Nasal swab and other specimen for local assay collected during assessment visit

If during passive or active surveillance contact, the investigator/study staff assesses that an infant presents a potential RSV-RTI (refer to 9.2), a nasal swab and another specimen for local assay will be collected during an assessment visit:

- The nasal swab will allow assessing the potential RSV infection by quantitative RT-PCR at sponsor laboratory.
- The specimen for local assay will allow assessing the potential RSV infection and RVP at a local routine laboratory, where available. The specimen type will depend on the assay run locally.

Nasal swab collected to detect asymptomatic RSV infections

During RSV season, there will be monthly nasal swabs to detect asymptomatic RSV infections. These nasal swabs will allow assessing the potential RSV infection by quantitative RT-PCR at sponsor laboratory.

Refer to the SPM for more details about nasal swab.

6.6.12. Detection of spontaneous bleeding or easy bruising and evidence of bruising or petechiae

Subjects' parent(s)/LAR(s) will be instructed to contact the investigator/study staff if their child presents symptoms of spontaneous bleeding or easy bruising or if their child develops a rash in the month following vaccination, in order to detect any thrombocytopenic petechiae or purpura. Refer to Sections 9.1.5 and 9.4.3 for reporting of adverse events of specific interest (spontaneous or excessive bleeding).

At Visit 2 (Day 1), Visit 3 (Day 3)*, Visit 4 (Day 7), Visit 6 (Day 31), Visit 7 (Day 33)*, and Visit 8 (Day 37), the investigator will perform a clinical examination with special attention given to detection of spontaneous bleeding or easy bruising and evidence of bruising or petechiae. Episodes of spontaneous bleeding or easy bruising and evidence of bruising or petechiae will be recorded in the eCRF.

* The visits on Day 3 and Day 33 may be replaced by a phone call by the investigator/study staff to inquire of the infants' general health and to solicit reports of a rash, spontaneous bleeding or easy bruising.

Further testing may be required to investigate a finding or guide subject management based on the investigators clinical judgment.

6.6.13. Detection of petechiae or evidence of petechiae induced by the tourniquet

At Visit 2 (Day 1), Visit 6 (Day 31), the investigator will perform an examination of the limb where the tourniquet for blood sample has been placed in order to detect petechiae or evidence of petechiae induced by the tourniquet. This special attention to detection of petechiae or evidence of petechiae induced by the tourniquet will also be given at Visit 4 (Day 7) and/or Visit 8 (Day 37) in case hematology testing will be performed on that day. Episodes of petechiae will be recorded in the eCRF.

These episodes might be reported as AE based on the investigators clinical judgment.

6.6.14. Surveillance for RSV-RTI and wheezing

Surveillance period for RSV-RTI and wheezing will start at Visit 1 (after Dose 1) until the final visit (Visit 11 [Day 730]). In order to timely detect RSV-RTI and to ensure cases are timely captured by the study sites, both active and passive surveillance will be conducted as described in Section 9.2. Contacts for active and passive surveillance will be recorded in the eCRF.

If an assessment visit is performed (refer to Section 9.2.4), this visit will be recorded in the eCRF, as well as signs and symptoms. Signs and symptoms to be recorded in the eCRF are listed in the SPM.

At the first vaccination visit (Visit 1 [Day 0]), RTI episode cards will be provided to the subject's parent(s)/LAR(s). At subsequent study visits, new RTI episode card(s) will be distributed as needed. The subject's parent(s)/LAR(s) will record the start date and the end date of the following symptoms: cough, runny nose, blocked nose, difficulty in breathing, or wheezing. The subject's parent(s)/LAR(s) will be instructed to return the completed RTI episode card to the investigator at the next visit or by mail (e-mail or postal mail). The investigator will transcribe the collected information into the eCRF. Any unreturned RTI episode card will be sought from the subject's parent(s)/LAR(s) through telephone call(s) or any other convenient procedure.

6.6.15. Check and record concomitant medication/vaccination

Concomitant medication/vaccination must be checked and recorded in the eCRF as described in Section 7.6.

6.6.16. Recording of AEs, AEs of specific interest and SAEs

- Refer to Section 9.3 for procedures for the investigator to record AEs, AEs of specific interest (RSV-LRTI and spontaneous or excessive bleeding) and SAEs.
- Refer to Section 9.4 for guidelines and how to report AEs of specific interest and SAE reports to GSK Biologicals.

- The subjects' parent(s)/LAR(s) will be instructed to contact the investigator immediately should the subjects manifest any signs or symptoms they perceive as serious.
- At each vaccination visits (Visit 1 [Day 0] and Visit 5 [Day 30]), a diary card will be provided to the subject's parent(s)/LAR(s). The subject's parent(s)/LAR(s) will record body temperature, any solicited local/general AEs, any unsolicited AEs, and concomitant medications/products/vaccinations on the day of vaccination and 6 subsequent days. The subject's parent(s)/LAR(s) will be instructed to return the completed diary card to the investigator at Visit 4 (Day 7) and Visit 8 (Day 37).

  The subject's parent(s)/LAR(s) will record on the same diary card any unsolicited AEs and concomitant medications/products/vaccinations from 7 to 29 days after each vaccination. The subject's parent(s)/LAR(s) will be instructed to return the completed diary card to the investigator at Visit 5 (Day 30) and Visit 9 (Day 60).

  The investigator will collect and verify completed diary cards during discussion with the subject's parent(s)/LAR(s) on Visit 4 (Day 7), Visit 5 (Day 30), Visit 8 (Day 37) and Visit 9 (Day 60). The investigator will transcribe the collected information into the eCRF in English.

  Any unreturned diary cards will be sought from the subject's parent(s)/LAR(s) through telephone call(s) or any other convenient procedure.

6.6.17. Screening conclusion

At the end of the screening visit, the investigator will:

- Review data collected.
- Complete the Screening Conclusion screen in the eCRF.

6.6.18. Signing of investigator signature form by investigator

At the end of the screening visit, Visit 9 (Day 60), Visit 10 (Day 365) and Visit 11 (Day 730), the investigator will sign the investigator signature form in the eCRF.

6.6.19. Study conclusion

At the last visit (Visit 11 [Day 730]), the investigator will:

- Review data collected to ensure accuracy and completeness.
- Complete the Study Conclusion screen in the eCRF.

6.7. Biological sample handling and analysis

Please refer to the SPM for details on biospecimen management (handling, storage and shipment).

Samples will not be labeled with information that directly identifies the subject but will be coded with the identification number for the subject (subject number).

- Collected samples will be used for protocol mandated research and purposes related to the improvement, development and quality assurance of the laboratory tests described in this protocol. This may include the management of the quality of these tests, the maintenance or improvement of these tests, the development of new test methods, as well as making sure that new tests are comparable to previous methods and work reliably.
- It is also possible that future findings may make it desirable to use the samples acquired in this study for future research, not described in this protocol. Therefore, all subjects in countries where this is allowed, will be asked to give a specific consent to allow GSK or a contracted partner to use the samples for future research. Future research will be subject to the laws and regulations in the respective countries and will only be performed once an independent Ethics Committee or Review Board has approved this research.

Information on further investigations and their rationale can be obtained from GSK Biologicals.

Any sample testing will be done in line with the consent of the individual subject's parent(s)/LAR(s).

Refer also to the Investigator Agreement, where it is noted that the investigator cannot perform any other biological assays except those described in the protocol or its amendment(s).

If additional testing is performed, the marker priority ranking given in Section 6.7.4 may be changed.

Collected samples will be stored for a maximum of 20 years (counting from when the last subject performed the last study visit), unless local rules, regulations or guidelines require different timeframes or different procedures, which will then be in line with the subject consent. These extra requirements need to be communicated formally to and discussed and agreed with GSK Biologicals.

6.7.1. Use of specified study materials

When materials are provided by GSK Biologicals, it is MANDATORY that all clinical samples (including serum samples) be collected and stored exclusively using those materials in the appropriate manner. The use of other materials could result in the exclusion of the subject from the ATP analysis (See Section 11.5 for the definition of cohorts to be analyzed). The investigator must ensure that his/her personnel and the

laboratory(ies) under his/her supervision comply with this requirement. However, when GSK Biologicals does not provide material for collecting and storing clinical samples, appropriate materials from the investigator's site must be used. Refer to the Module on Clinical Trial Supplies in the SPM.

6.7.2. Biological samples

Table 7 Biological samples (Amended 10 December 2017)

Sample	Timepoint	Type of sample†	Subject	Number of subjects§	Quantity
Blood	Screening	Blood sample for RSV serostatus	All infants	≥ 96	1.0 mL
		Blood sample for hematology	All infants	≥ 96	1.2 mL
		Blood sample for biochemistry	All infants	≥ 96	1.1 mL
		Blood sample for cell-mediated immunogenicity response #	All infants††	≥ or≤ 96 ††,¥	2.0 mL
		Blood sample for humoral response #	All infants	≥ 96¥	2.5 mL
		Total volume of blood collected for each subject††			**7.8 mL**
	Visit 2 (Day 1)	Blood sample for hematology	All infants	96	1.2 mL
		Total volume of blood collected for each subject			**1.2 mL**
	Visit 4 (Day 7)	Blood sample for hematology	All infants‡	≤96‡	1.2 mL
		Total volume of blood collected for each subject‡			**1.2 mL**
	Visit 5 (Day 30)	Blood sample for hematology	All infants	96	1.2 mL
		Blood sample for biochemistry	All infants	96	1.1 mL
		Blood sample for cell-mediated immunogenicity response	All infants††	≤ 96††	2.0 mL
		Blood sample for humoral response	All infants	96	2.5 mL
		Total volume of blood collected for each subject††			**6.8 mL**
	Visit 6 (Day 31)	Blood sample for hematology	All infants	96	1.2 mL
		Total volume of blood collected for each subject			**1.2 mL**
	Visit 8 (Day 37)	Blood sample for hematology	All infants‡	≤96‡	1.2 mL
		Total volume of blood collected for each subject‡			**1.2 mL**
	Visit 9 (Day 60)	Blood sample for hematology	All infants	96	1.2 mL
		Blood sample for biochemistry	All infants	96	1.1 mL
		Blood sample for cell-mediated immunogenicity response	All infants††	≤ 96††	2.0 mL
		Blood sample for humoral response	All infants	96	2.5 mL
		Total volume of blood collected for each subject††			**6.8 mL**
	Total volume of blood collected for each subject††,‡ from Screening to Visit 9 (Day 60)*				**26.2 mL**
	Visit 10 (Day 365)	Blood sample for cell-mediated immunogenicity response	All infants††	≤ 96††	2.0 mL
		Blood sample for humoral response	All infants	96	2.5 mL
		Total volume of blood collected for each subject††			**4.5 mL**
	Assessment of potential RSV-RTI	Blood sample for assessment of mechanism of illness (potential ERD)	**Hospitalization for RSV-LRTI**	**Hospitalization for RSV-LRTI**	**2.5 mL**
	Unscheduled visit for safety	Blood sample for hematology	Event-driven	Event-driven	1.2 mL
		Blood sample for biochemistry	Event-driven	Event-driven	1.1 mL
	Total volume of blood collected for each subject††,‡ from Screening to Visit 11 (Day 730)*				**30.7 mL**

Sample	Timepoint	Type of sample†	Subject	Number of subjects§	Quantity
Nasal swab	Assessment of potential RSV-RTI	Nasal swab**	Event-driven	Event-driven	-
	Surveillance for asymptomatic RSV-RTI	Nasal swab***	All infants	96	-

§ The target will be to enroll up to 96 RSV-seropositive infants aged 12 to 23 months at the time of first vaccination (unless the dose escalation IDMC decision can be made based on the evaluation of accumulating safety data on fewer subjects) to ensure that at least 64 infants receive two doses of study vaccine (ChAd155-RSV or Placebo) (see Section 5.1) **(Amended 10 December 2017).**

* Total quantity of blood for each subject excludes any unscheduled hematology and biochemistry blood sampling or blood sample for assessment of mechanism of illness collected if a subject has been hospitalized for RSV-LRTI.

** If during passive or active surveillance contact, the investigator/study staff assesses that an infant presents a potential RSV-RTI (refer to Table 4 for definition), the potential RSV infection will be assessed by quantitative RT-PCR of nasal swab specimens taken at an assessment visit.

*** During RSV season, there will be monthly nasal swabs to detect asymptomatic RSV infections. These swabs will be tested by quantitative RT-PCR at sponsor laboratory.

These assays will be performed only on enrolled infants.

¥ Blood samples for CMI and for humoral immunogenicity may be postponed until after the RSV serostatus is known and eligibility is confirmed up to or on Day 0 (but before vaccination).

† The priority ranking in blood sampling is hematology > biochemistry > humoral response > CMI response. Venipuncture will not be repeated for CMI and humoral response in case of insufficient volume, at any timepoint.

†† In investigational sites with a laboratory in proximity capable to perform WBS necessary for the CMI assay.

‡ Day 7 hematology testing will be performed if any ≥ Grade 1 for platelet decrease is detected on Day 1 after vaccination. Similarly, hematology testing on Day 37 will be performed if any ≥ Grade 1 for platelet decrease is detected on Day 31.

6.7.3. Laboratory assays

Please refer to Appendix A for a detailed description of the assays performed in the study. Please refer to Appendix B for the address of the clinical laboratories planned to be used for sample analysis.

The following laboratory assays are planned:

- Frequency of interleukin 2, 13, and 17 (IL-2, IL-13 and IL-17), cluster of differentiation 40 ligand (CD40L), 41BB, tumor necrosis factor alpha (TNF-α), and IFN-γ secreting CD3+/CD4+ and CD3+/CD8+ T-cells will be determined by ICS assay on whole blood samples (Table 8).
- Serostatus to RSV F and/or G protein will be determined at screening by ELISA commercial kit on serum sample, according to the manufacturer instructions (Table 9).
- Functional (neutralizing) antibody titers against RSV-A will be measured by a neutralization assay on serum samples (Table 9).
- RSV F protein antibody concentrations and palivizumab-competing antibody concentrations will be determined by in-house ELISA on serum sample (Table 9).

- RTI will be assessed by:
  - Quantitative RT-PCR that is able to discriminate RSV-A and RSV-B subtypes (Table 10).
  - Qualitative multiplex PCR for detection of a panel of viruses (Table 10).
  - Local RSV assay and local RVP where available (Table 10).

These assays will be performed at the investigator's laboratory, at GSK Biologicals' laboratory or at laboratory designated by GSK Biologicals using standardized and validated procedures.

Table 8 Cell-Mediated Immunity (CMI)

System	Component	Challenge	Method	Unit	Laboratory
Whole blood	IL-2, CD40L, 41BB, TNF-α, IFN-γ, IL-13, IL-17 secreting CD3+/CD4+ and CD3+/CD8+ T-cells	Peptide pools covering the proteins F, N & M2-1	ICS	Events/10E6 cells	GSK Biologicals[1] or designated laboratory

CD40L: cluster of differentiation 40 ligand; **ICS**: Intracellular staining; **IFN-γ**: interferon gamma, **IL (IL-2; IL-13; IL-17)**: interleukin; **TNF-α**: tumor necrosis factor alpha
[1] GSK Biologicals laboratory refers to the clinical laboratory sciences (CLS) in Rixensart, Belgium; Wavre, Belgium.

Table 9 Humoral Immunity (Antibody determination)

System	Component	Method	Kit / Manufacturer	Unit	Cut-off	Laboratory
SERUM	Respiratory Syncytial virus Ab.IgG (RSV F or G antibody)	ELISA	IBL International	Qualitative assay (positive/negative)	Qualitative assay (positive/negative)	Q[2] Solutions
SERUM	Respiratory Syncytial Virus A Ab	NEUTRALIZATION	In-house	ED60	To be defined	GSK Biologicals[1] or NÉOMED-LABS
SERUM	Respiratory Syncytial Virus F protein Ab.IgG (Anti-RSV PreF antibody)	ELISA	In-house	EU/ml	To be defined	GSK Biologicals[1] or NÉOMED-LABS
SERUM	Respiratory Syncytial Virus F protein Ab.IgG (Palivizumab competing Ab)	ELISA	In-house	µg/ml	To be defined	GSK Biologicals[1] or NÉOMED-LABS

Ab: antibody; **ELISA**: enzyme-linked immunosorbent assay; **IgG**: immunoglobulin G; **RSV**: respiratory syncytial virus
[1] GSK Biologicals laboratory refers to the CLS in Rixensart, Belgium; Wavre, Belgium.

Table 10 Molecular Biology (PCR tests)

System	Component	Kit / Manufacturer	Method	Unit	Laboratory
Nasal swab	RSV A RNA RSV B RNA	In-house	Quantitative RT-PCR	Copies/ml	GSK Biologicals[1] or designated laboratory
Nasal swab	Influenza A virus (Flu A) Influenza B virus (Flu B) Human respiratory syncytial virus A (RSV A) Human respiratory syncytial virus B (RSV B) Human Influenza A virus subtype H1 (Flu A-H1) Human Influenza A virus subtype H3 (Flu A-H3) Human Influenza A virus subtype H1pdm09 (Flu A-H1pdm09) Human adenovirus (AdV) Human metapneumovirus (MPV) Human enterovirus (HEV) Human parainfluenza virus 1 (PIV1) Human parainfluenza virus 2 (PIV2) Human parainfluenza virus 3 (PIV3) Human parainfluenza virus 4 (PIV4) Human bocavirus 1/2/3/4 (HBoV) Human rhinovirus A/B/C (HRV) Human coronavirus 229E (229E) Human coronavirus NL63 (NL63) Human coronavirus OC43 (OC43) [2]	Allplex Respiratory Panel or equivalent [2]	Multiplex PCR	Qualitative assay (positive/ negative)	GSK Biologicals[1] or designated laboratory
Other specimen	RSV	Not applicable	Locally available RSV assay	Not applicable	Routine local laboratory
Other specimen	RVP	Not applicable	Locally available RVP assay	Not applicable	Routine local laboratory

PCR: polymerase chain reaction; **RSV**: respiratory syncytial virus; **RT:** Reverse Transcription; **RVP**: respiratory viral panel

[1] GSK Biologicals laboratory refers to the CLS in Rixensart, Belgium; Wavre, Belgium.

[2] The list of components might be subject to change in case equivalent kit is used for multiplex RVP testing.

Additional testing on serum and blood samples to characterize the immune response to RSV or to the investigational ChAd155-RSV vaccine may be performed if deemed necessary for accurate interpretation of the data and/or should such test(s) become available in the GSK Biologicals laboratory or a laboratory designated by GSK Biologicals.

Additional viral diagnosis testing on the nasal swabs, such as (but not limited to) multiplex PCR and/or high-throughput sequencing, may be done, if deemed necessary, provided specific assays become available at GSK Biologicals' laboratory or a laboratory designated by GSK Biologicals.

Additional testing may include, but is not limited to, the following:

- Anti-vector immunity: neutralization.

For subjects hospitalized for LRTI and RSV-positive using a locally available RSV test (refer to Section 9.2.5), an additional blood sample will be collected. The corresponding serum sample will be stored at GSK Biologicals laboratory to further characterize the mechanisms of illness (potential ERD), if deemed necessary. In this event, the serum samples could be tested by RSV neutralization assay and also by ELISA (or alternative method) for measurement of protein F antibodies and cytokines.

Hematology and biochemistry assessments will be performed in the investigator's laboratory as per local practice using standardized and validated procedures (Table 11).

Table 11 Hematology and biochemistry tests (Amended 10 December 2017)

System	Discipline	Component	Timepoint	Method	Scale	Laboratory
Whole blood	Hematology	Hemoglobin	Screening, Day 1, Day 7*, Day 30, Day 31, Day 37*, Day 60	As per local practice	Quantitative	Local laboratory
		Leukocytes (White Blood Cells)**				
		Platelets				
Serum	Biochemistry	Alanine Aminotransferase	Screening, Day 30, Day 60	As per local practice	Quantitative	
		Aspartate Aminotransferase				
		Creatinine				

* Day 7 hematology testing will be performed if any ≥ Grade 1 for platelet decrease is detected on Day 1 after vaccination. Similarly, hematology testing on Day 37 will be performed if any ≥ Grade 1 for platelet decrease is detected on Day 31.

** ***White Blood Cell differential including absolute neutrophil and lymphocyte counts must be recorded in the eCRF.*** **(Amended 10 December 2017)**

Additional hematology and/or biochemistry testing may be performed or required in specific circumstances. Refer to Figure 2 for a schematic overview.

Collected samples may be used for purposes related to the quality assurance of the laboratory tests described in this protocol. This may include the management of the quality of these current tests, the maintenance or improvement of these current tests, the development of new test methods for the markers described in this protocol, as well as making sure that new tests are comparable to previous methods and work reliably.

The GSK Biologicals' clinical laboratories have established a Quality System supported by procedures. The activities of GSK Biologicals' clinical laboratories are audited regularly for quality assessment by an internal (sponsor-dependent) but laboratory-independent Quality Department.

6.7.4. Biological samples evaluation

6.7.4.1. Immunological read-outs

Table 12 Immunological read-outs (Amended 10 December 2017)

Blood sampling timepoint			No. subjects†	Component	Components priority rank
Type of contact and timepoint*	Sampling timepoint	System			
Screening (Day -29 to Day 0)	Pre-Vaccination	Whole blood	≤96‡,¥	IL-2, CD40L, 41BB, TNF-α, IFN-γ, IL-13, IL-17 secreting T-cells	1
		Serum	≥ 96	Respiratory Syncytial virus Ab.IgG (RSV F or G antibody)	1
			≥96¥	Anti-RSV A Neutralizing Antibody	2
				Respiratory Syncytial Virus F protein Ab.IgG (Anti-RSV PreF antibody)	3
				Respiratory Syncytial Virus F protein Ab.IgG (Palivizumab competing Ab)	4
Visit 5 (Day 30)	Post-Vaccination 1	Whole blood	≤96‡	IL-2, CD40L, 41BB, TNF-α, IFN-γ, IL-13, IL-17 secreting T-cells	1
		Serum	~96	Anti-RSV A Neutralizing Antibody	1
				Respiratory Syncytial Virus F protein Ab.IgG (Anti-RSV PreF antibody)	2
				Respiratory Syncytial Virus F protein Ab.IgG (Palivizumab competing Ab)	3
Visit 9 (Day 60)	Post-Vaccination 2	Whole blood	≤96‡	IL-2, CD40L, 41BB, TNF-α, IFN-γ, IL-13, IL-17 secreting T-cells	1
		Serum	~96	Anti-RSV A Neutralizing Antibody	1
				Respiratory Syncytial Virus F protein Ab.IgG (Anti-RSV PreF antibody)	2
				Respiratory Syncytial Virus F protein Ab.IgG (Palivizumab competing Ab)	3
Visit 10 (Day 365)	Post-Vaccination 2	Whole blood	≤96‡	IL-2, CD40L, 41BB, TNF-α, IFN-γ, IL-13, IL-17 secreting T-cells	1
		Serum	~96	Anti-RSV A Neutralizing Antibody	1
				Respiratory Syncytial Virus F protein Ab.IgG (Anti-RSV PreF antibody)	2

CD40L: cluster of differentiation 40 ligand; **IFN-γ**: interferon gamma; **IgG**: immunoglobulin G; **IL (IL-2; IL-13; IL-17)**: interleukin; **TNF-α**: tumor necrosis factor alpha; **RSV**: respiratory syncytial virus.

* Blood samples for CMI taken on Day 7 and Day 37 for subjects enrolled under protocol amendment 1 will still be assessed for CMI.

† ***The target will be to enroll up to 96 RSV-seropositive infants aged 12 to 23 months at the time of first vaccination (unless the dose escalation IDMC decision can be made based on the evaluation of accumulating safety data on fewer subjects) to ensure that at least 64 infants receive two doses of study vaccine (ChAd155-RSV or Placebo) (see Section 5.1)*** **(Amended 10 December 2017).**

‡ Only in investigational sites with a laboratory in proximity capable to perform WBS necessary for the CMI assay.

¥ Blood samples for CMI and for humoral immunogenicity may be postponed until after the RSV serostatus is known and eligibility is confirmed up to or on Day 0(but before vaccination).

In case of insufficient blood sample volume to perform assays for all antibodies, the samples will be analyzed according to priority ranking provided in Table 12.

Table 13 Molecular biology for nasal swab and specimen analysis (Amended 10 December 2017)

Blood sampling timepoint			No. subjects†	Component	Components priority rank
Type of contact and timepoint	Sampling timepoint	System			
Surveillance for asymptomatic RSV-RTI	Monthly contact during the RSV season	Nasal swab	~96	Quantitative RT-PCR (RSV A RNA and RSV B RNA)	Not applicable
Assessment of potential RSV-RTI	All subjects attending assessment visit	Nasal swab	Event-driven	Quantitative RT-PCR (RSV A RNA and RSV B RNA)	Not applicable
Assessment of potential RSV-RTI	All subjects attending assessment visit with positive Quantitative RSV A/B RT-PCR results	Nasal swab	Event-driven	Qualitative multiplex PCR** (Influenza A virus (Flu A) Influenza B virus (Flu B) Human respiratory syncytial virus A (RSV A) Human respiratory syncytial virus B (RSV B) Human Influenza A virus subtype H1 (Flu A-H1) Human Influenza A virus subtype H3 (Flu A-H3) Human Influenza A virus subtype H1pdm09 (Flu A-H1pdm09) Human adenovirus (AdV) Human metapneumovirus (MPV) Human enterovirus (HEV) Human parainfluenza virus 1 (PIV1) Human parainfluenza virus 2 (PIV2) Human parainfluenza virus 3 (PIV3) Human parainfluenza virus 4 (PIV4) Human bocavirus 1/2/3/4 (HBoV) Human rhinovirus A/B/C (HRV) Human coronavirus 229E (229E) Human coronavirus NL63 (NL63) Human coronavirus OC43 (OC43))	Not applicable
Assessment of potential RSV-RTI	All subjects presenting confirmed LRTI* (with negative Quantitative RSV A and B RT-PCR results)	Nasal swab	Event-driven	Qualitative multiplex PCR** (Influenza A virus (Flu A) Influenza B virus (Flu B) Human respiratory syncytial virus A (RSV A) Human respiratory syncytial virus B (RSV B) Human Influenza A virus subtype H1 (Flu A-H1) Human Influenza A virus subtype H3 (Flu A-H3) Human Influenza A virus subtype H1pdm09 (Flu A-H1pdm09) Human adenovirus (AdV) Human metapneumovirus (MPV) Human enterovirus (HEV) Human parainfluenza virus 1 (PIV1) Human parainfluenza virus 2 (PIV2) Human parainfluenza virus 3 (PIV3) Human parainfluenza virus 4 (PIV4) Human bocavirus 1/2/3/4 (HBoV) Human rhinovirus A/B/C (HRV) Human coronavirus 229E (229E) Human coronavirus NL63 (NL63) Human coronavirus OC43 (OC43))	Not applicable

Blood sampling timepoint			No. subjects†	Component	Components priority rank
Type of contact and timepoint	Sampling timepoint	System			
Assessment of potential RSV-RTI	All subjects attending assessment visit	Other specimen	Event-driven	Locally available RSV assay	Not applicable
Assessment of potential RSV-RTI	All subjects attending assessment visit	Other specimen	Event-driven	Locally available RVP assay	Not applicable

PCR: polymerase chain reaction; **RSV**: respiratory syncytial virus; **RTI**: respiratory tract infection; **RVP**: respiratory viral panel.

† ***The target will be to enroll up to 96 RSV-seropositive infants aged 12 to 23 months at the time of first vaccination (unless the dose escalation IDMC decision can be made based on the evaluation of accumulating safety data on fewer subjects) to ensure that at least 64 infants receive two doses of study vaccine (ChAd155-RSV or Placebo) (see Section 5.1)*** **(Amended 10 December 2017).**

* Refer to Table 4 for the case definitions.

** The list of components might be subject to change in case equivalent kit is used for multiplex RVP testing.

6.7.5. Immunological correlates of protection

No generally accepted immunological correlate of protection has been established so far for the antigens used in the candidate ChAd155-RSV vaccine.

7. STUDY VACCINES AND ADMINISTRATION

7.1. Description of study vaccines

The investigational ChAd155-RSV vaccine has been developed and manufactured by GSK Biologicals.

The Quality Control Standards and Requirements for the candidate vaccine are described in separate Quality Assurance documents (e.g. release protocols, certificate of analysis) and the required approvals have been obtained.

The vaccines are labeled and packed according to applicable regulatory requirements.

Table 14 Study vaccines

Treatment name	Vaccine name	Formulation	Presentation	Volume to be administered	Number of doses
ChAd155 $5x10^9$ vp	ChAd155-RSV	ChAd155-RSV=$1*10^{10}$vp/mL	Liquid in monodose vial	0.5 ml	2
ChAd155 $1.5x10^{10}$ vp	ChAd155-RSV	ChAd155-RSV=$1*10^{11}$vp/mL	Liquid in monodose vial	0.15 ml	2
ChAd155 $5x10^{10}$ vp	ChAd155-RSV	ChAd155-RSV=$1*10^{11}$vp/mL	Liquid in monodose vial	0.5 ml	2
Placebo 0.5	Formulation buffer S9b	Na_2HPO_4=1.3mg; KH_2PO_4=186µg; NaCl=3.85mg; KCl=100µg; $MgCl_2$=50µg *	Clear liquid	0.5 ml	2
Placebo 0.15	Formulation buffer S9b	Na_2HPO_4=0.4mg; KH_2PO_4=56µg; NaCl=1,16mg; KCl=30µg; $MgCl_2$=15µg *	Clear liquid	0.15 ml	2

* Same concentration of buffer S9b is used for Placebo 0.5 and Placebo 0.15. Values indicated here are the quantities contained in the volume to be administered.

7.2. Storage and handling of study vaccines

The study vaccine must be stored at the respective label storage temperature conditions in a safe and locked place. Access to the storage space should be limited to authorized study personnel. The storage conditions will be assessed during pre-study activities under the responsibility of the sponsor study contact. The storage temperature should be continuously monitored with calibrated (if not validated) temperature monitoring device(s) and recorded. Refer to the Module on Clinical Trial Supplies in the SPM for more details on storage of the study vaccines.

Temperature excursions must be reported in degree Celsius.

For the placebo any temperature excursion outside the range of 0.0 to +8.0°C (for +2 to +8°C/+36 to +46°F label storage condition) and for the ChAd155-RSV vaccine any temperature excursion above -60°C (for ≤ -60°C/≤ -76°F label storage condition) or below -90°C must be reported in the appropriate (electronic) temperature excursion decision form ([e]TDF). The impacted IMPs must not be used and must be stored in quarantine at label temperature conditions until usage approval has been obtained from the sponsor.

Refer to the Module on Clinical Trial Supplies in the SPM for details and instructions on the temperature excursion reporting and usage decision process, packaging and accountability of the study vaccines.

7.3. Dosage and administration of study vaccines

Table 15 Dosage and administration

Type of contact and timepoint	Volume to be administered	Study group	Treatment name	Route[1]	Site	Side[3]
Visit 1 (Day 0) Visit 5 (Day 30)	0.5 ml	RSV-Ld	ChAd155 $5x10^9$ vp	IM	Deltoid[2]	Left
	0.5 ml	Placebo-Ld	Placebo 0.5	IM	Deltoid[2]	Left
	0.15 ml	RSV-Md	ChAd155 $1.5x10^{10}$ vp	IM	Deltoid[2]	Left
	0.15 ml	Placebo-Md	Placebo 0.15	IM	Deltoid[2]	Left
	0.5 ml	RSV-Hd	ChAd155 $5x10^{10}$ vp	IM	Deltoid[2]	Left
	0.5 ml	Placebo-Hd	Placebo 0.5	IM	Deltoid[2]	Left

[1] **IM**: Intramuscular.
[2] If the muscle size is adequate; otherwise injections might be done in the anterolateral thigh. The site of administration of Dose 2 should preferably remain the same as the site of administration of Dose 1.
[3] Preferable side for administration.
RSV-Ld: low dose ChAd155-RSV vaccine (5×10^9 vp); **RSV-Md**: middle dose ChAd155-RSV vaccine (1.5×10^{10} vp); **RSV-Hd**: high dose ChAd155-RSV vaccine (5×10^{10} vp).

Note that ChAd155-RSV vaccine middle dose will be given as a 0.15-mL fractional dose of the high dose (0.5 mL). Refer to the SPM for detailed instructions on study vaccine administration.

Minimizing environmental contamination with genetically modified organisms

Each product will be used in accordance with the applicable genetically modified organism regulations.

To minimize release of the recombinant vectored vaccine virus into the environment, each vaccine is produced under good manufacturing practice conditions with the handling of live material in appropriate laboratory facilities. This is to ensure that any release of modified organism is contained, inactivated and incinerated, using single use equipment as much as possible, to avoid release of modified genetic material into the environment.

After each vaccination, the injection site will be covered with a dressing in order to absorb any virus that may leak out through the needle track. The dressing will be removed only after 30 minutes and will be disposed as genetically modified organism waste by autoclaving or in accordance with the applicable guidelines/SOPs at the investigator's site.

7.4. Replacement of unusable vaccine doses

In addition to the vaccine doses provided for the planned number of subjects (including over-randomization when applicable), at least 10% additional vaccine doses will be supplied to replace those that are unusable.

The investigator will use SBIR to obtain the replacement vial number. The replacement numbers will be allocated by dose. The system will ensure, in a blinded manner, that the replacement vial matches the formulation the subject was assigned to by randomization.

7.5. Contraindications to subsequent vaccination

The following events constitute absolute contraindications to further administration of ChAd155-RSV vaccine. If any of these events occur during the study, the subject must not receive additional doses of vaccine but may continue other study procedures at the discretion of the investigator (see Section 9.5).

- Anaphylaxis following the administration of vaccine.
- Any confirmed or suspected immunosuppressive or immunodeficient condition, including HIV infection.
- Any condition that in the judgment of the investigator would make IM injection unsafe.
- Current autoimmune disease.
- Acute or chronic, clinically significant pulmonary, cardiovascular, hepatic or renal functional abnormality, as determined by physical examination or laboratory tests.
- Failure to thrive.

The following events constitute contraindications to administration of ChAd155-RSV vaccine at that point in time; if any of these events occur at the time scheduled for vaccination, the subject may be vaccinated at a later date, within the time window specified in the protocol (see Section 6.5), or the subject may be withdrawn at the discretion of the investigator (see Section 9.5).

- Acute disease and/or fever at the time of vaccination.
  - Fever is defined as temperature ≥ 37.5°C/99.5°F for oral, axillary or tympanic route, or ≥ 38.0°C/100.4°F for rectal route. The preferred route for recording temperature in this study will be axillary.
  - Subjects with a minor illness (such as mild diarrhea, mild upper respiratory tract infections) without fever can be administered all vaccines.
- Any Grade 1 abnormality with potential clinical relevance or any ≥ Grade 2 abnormality in hematological parameters detected in the Day 7 blood sample (refer to Section 6.6.11.2, Figure 2 and Appendix C).

7.6. Concomitant medications/products and concomitant vaccinations

At each study visit, the investigator should question the subject's parent(s)/LAR(s) about any medications/products taken and vaccinations received by the subject.

7.6.1. Recording of concomitant medications/products and concomitant vaccinations

The following concomitant medication(s)/product(s)/vaccine(s) must be recorded in the eCRF.

- All concomitant medications/products, except vitamins and dietary supplements, administered in the 30 day follow-up period after the administration of each dose of study vaccines (Day 0 to Day 29).
- Any concomitant vaccination administered in the period starting 30 days before the first dose of study vaccines and ending at the last study visit (Day -29 to Day 730).
- Prophylactic medication (i.e. medication administered in the absence of ANY symptom and in anticipation of a reaction to the vaccination or blood sampling).
  - E.g. an anti-pyretic is considered to be prophylactic when it is given in the absence of fever and any other symptom, to prevent fever from occurring [fever is defined as temperature ≥ 37.5°C/99.5°F for oral, axillary or tympanic route, or ≥ 38.0°C/100.4°F for rectal route].
  - E.g. a topical local anesthetic used prior to a blood sampling is considered to be prophylactic.
- Any concomitant medications/products/vaccines listed in Section 7.6.2.
- Any concomitant medications/products/vaccines relevant to a SAE to be reported as per protocol or administered at any time during the study period for the treatment of a SAE. In addition, concomitant medications relevant to SAEs need to be recorded on the expedited Adverse Event report.
- Any antipyretic administered in the period starting 6 hours before vaccination and ending 12 hours after vaccination need to be recorded on the specific page of the eCRF.

7.6.2. Concomitant medications/products/vaccines that may lead to the elimination of a subject from ATP analyses

The use of the following concomitant medications/products/vaccines will not require withdrawal of the subject from the study but may determine a subject's evaluability in the ATP analysis. See Section 11.5 for cohorts to be analyzed.

- Any investigational or non-registered product (drug or vaccine) other than the study vaccine used during the study period.
- Immunosuppressants or other immune-modifying drugs administered chronically (i.e. more than 14 days in total) during the study period. For corticosteroids, this will mean prednisone ≥ 0.5 mg/kg/day (for pediatric subjects), or equivalent. Inhaled and topical steroids are allowed.
- Long-acting immune-modifying drugs administered at any time during the study period (e.g. infliximab).

- A vaccine not foreseen by the study protocol administered during the period starting 30 days before the first dose and ending 30 days after the last dose of vaccines administration*, with the exception of:
  - Scheduled routine pediatric vaccines which may be administered ≥ 14 days before a dose and ≥ 7 days after a dose of study vaccines.
  - Vaccines needed for urgent individual medical need (e.g. rabies prophylaxis).

  * In case an emergency mass vaccination for an unforeseen public health threat (e.g.: a pandemic) is organized by the public health authorities, outside the routine immunization program, the time period described above can be reduced if necessary for that vaccine provided it is licensed and used according to its Summary of Product Characteristics or Prescribing Information and according to the local governmental recommendations and provided a written approval of the sponsor is obtained.
- Immunoglobulins and/or any blood products administered during the study period.

8. HEALTH ECONOMICS

Not applicable.

9. SAFETY

The investigator or site staff is/are responsible for the detection, documentation and reporting of events meeting the criteria and definition of an AE or SAE as provided in this protocol.

Each subject's parent(s)/LAR(s) will be instructed to contact the investigator immediately should they/the subject manifest any signs or symptoms they perceive as serious.

9.1. Safety definitions

9.1.1. Definition of an adverse event

An AE is any untoward medical occurrence in a clinical investigation subject, temporally associated with the use of a medicinal product, whether or not considered related to the medicinal product.

An AE can therefore be any unfavorable and unintended sign (including an abnormal laboratory finding), symptom, or disease (new or exacerbated) temporally associated with the use of a medicinal product. For marketed medicinal products, this also includes failure to produce expected benefits (i.e. lack of efficacy), abuse or misuse.

Examples of an AE include:

- Significant or unexpected worsening or exacerbation of the condition/indication under study.

- Exacerbation of a chronic or intermittent pre-existing condition including either an increase in frequency and/or intensity of the condition.
- New conditions detected or diagnosed after investigational vaccine(s) administration even though they may have been present prior to the start of the study.
- Signs, symptoms, or the clinical sequelae of a suspected interaction.
- Signs, symptoms, or the clinical sequelae of a suspected overdose of either investigational vaccine(s) or a concurrent medication (overdose per se should not be reported as an AE/SAE).
- Signs, symptoms temporally associated with vaccine(s) administration.
- Significant failure of expected pharmacological or biological action.

AEs to be recorded as endpoints (solicited AEs) are described in Section 9.1.3. All other AEs will be recorded as UNSOLICITED AEs.

Examples of an AE DO NOT include:

- Medical or surgical procedures (e.g. endoscopy, appendectomy); the condition that leads to the procedure is an AE/SAE.
- Situations where an untoward medical occurrence did not occur (e.g. social and/or convenience admission to a hospital, admission for routine examination).
- Anticipated day-to-day fluctuations of pre-existing disease(s) or condition(s) present or detected at the start of the study that do not worsen.
- Pre-existing conditions or signs and/or symptoms present in a subject prior to the first study vaccination. These events will be recorded in the medical history section of the eCRF.

9.1.2. Definition of a serious adverse event

A SAE is any untoward medical occurrence that:

a. Results in death,

b. Is life-threatening,

   Note: The term ‘life-threatening’ in the definition of ‘serious’ refers to an event in which the subject was at risk of death at the time of the event. It does not refer to an event, which hypothetically might have caused death, had it been more severe.

c. Requires hospitalization or prolongation of existing hospitalization,

   Note: In general, hospitalization signifies that the subject has been admitted at the hospital or emergency ward for observation and/or treatment that would not have been appropriate in the physician’s office or in an out-patient setting. Complications that occur during hospitalization are also considered AEs. If a complication prolongs hospitalization or fulfills any other serious criteria, the event will also be considered

serious. When in doubt as to whether 'hospitalization' occurred or was necessary, the AE should be considered serious.

Hospitalization for elective treatment of a pre-existing condition (known or diagnosed prior to informed consent signature) that did not worsen from baseline is NOT considered an AE.

d. Results in disability/incapacity

Note: The term disability means a substantial disruption of a person's ability to conduct normal life functions. This definition is not intended to include experiences of relatively minor medical significance such as uncomplicated headache, nausea, vomiting, diarrhea, influenza like illness, and accidental trauma (e.g. sprained ankle) which may interfere or prevent everyday life functions but do not constitute a substantial disruption.

Medical or scientific judgment should be exercised in deciding whether reporting is appropriate in other situations, such as important medical events that may not be immediately life-threatening or result in death or hospitalization but may jeopardize the subject or may require medical or surgical intervention to prevent one of the other outcomes listed in the above definition. These should also be considered serious. Examples of such events are invasive or malignant cancers, intensive treatment in an emergency room or at home for allergic bronchospasm, blood dyscrasias or convulsions that do not result in hospitalization.

9.1.3. Solicited adverse events

9.1.3.1. Solicited local (injection-site) adverse events

The following local (injection-site) AEs will be solicited:

Table 16 Solicited local adverse events

All age groups
Pain at injection site
Redness at injection site
Swelling at injection site

9.1.3.2. Solicited general adverse events

The following general AEs will be solicited:

Table 17 Solicited general adverse events

Infant
Drowsiness
Fever*
Irritability/Fussiness
Loss of appetite

* Fever is defined as temperature ≥ 37.5°C/99.5°F for oral, axillary or tympanic route, or ≥ 38.0°C/100.4°F for rectal route. The preferred route for recording temperature in this study will be axillary.

Note: Temperature will be recorded in the evening. Should additional temperature measurements be performed at other times of day, the highest temperature will be recorded in the eCRF.

9.1.4. Clinical laboratory parameters and other abnormal assessments qualifying as adverse events or serious adverse events

In absence of diagnosis, abnormal laboratory findings (e.g. clinical chemistry, hematology, urinalysis) or other abnormal assessments (e.g. physical examination) that are judged by the investigator to be clinically significant will be recorded as AE or SAE if they meet the definition of an AE or SAE (refer to Sections 9.1.1 and 9.1.2). Clinically significant abnormal laboratory findings or other abnormal assessments that are present at baseline and significantly worsen following the start of the study will also be reported as AEs or SAEs.

The investigator will exercise his or her medical and scientific judgment in deciding whether an abnormal laboratory finding or other abnormal assessment is clinically significant.

9.1.5. Adverse events of specific interest

9.1.5.1. Spontaneous or excessive bleeding

Any episode of spontaneous or excessive bleeding occurring within 30 days after each vaccination should be fully investigated with a full range of hematological tests to identify the underlying cause and reported as an AE of specific interest.

9.1.5.2. RSV-LRTI

All subjects experiencing a LRTI associated with RSV infection should be reported as AE of specific interest in an expedited manner for the duration of their participation to the study. These will be detected through the surveillance for RSV-RTI (refer to Section 9.2).

To identify RSV-LRTI for the purpose of AE of specific interest, the diagnosis should be based on the investigators' clinical judgment taking into account the clinical history, the examination, relevant medical investigations and locally-available diagnostic test for RSV. Of note the case definition presented in Table 4 is based on a limited set of symptoms and signs and is designed for protocol specified analyses pooled across sites at study conclusion and should not be used for the purpose of reporting RSV-LRTI as an AE of specific interest during the trial conduct.

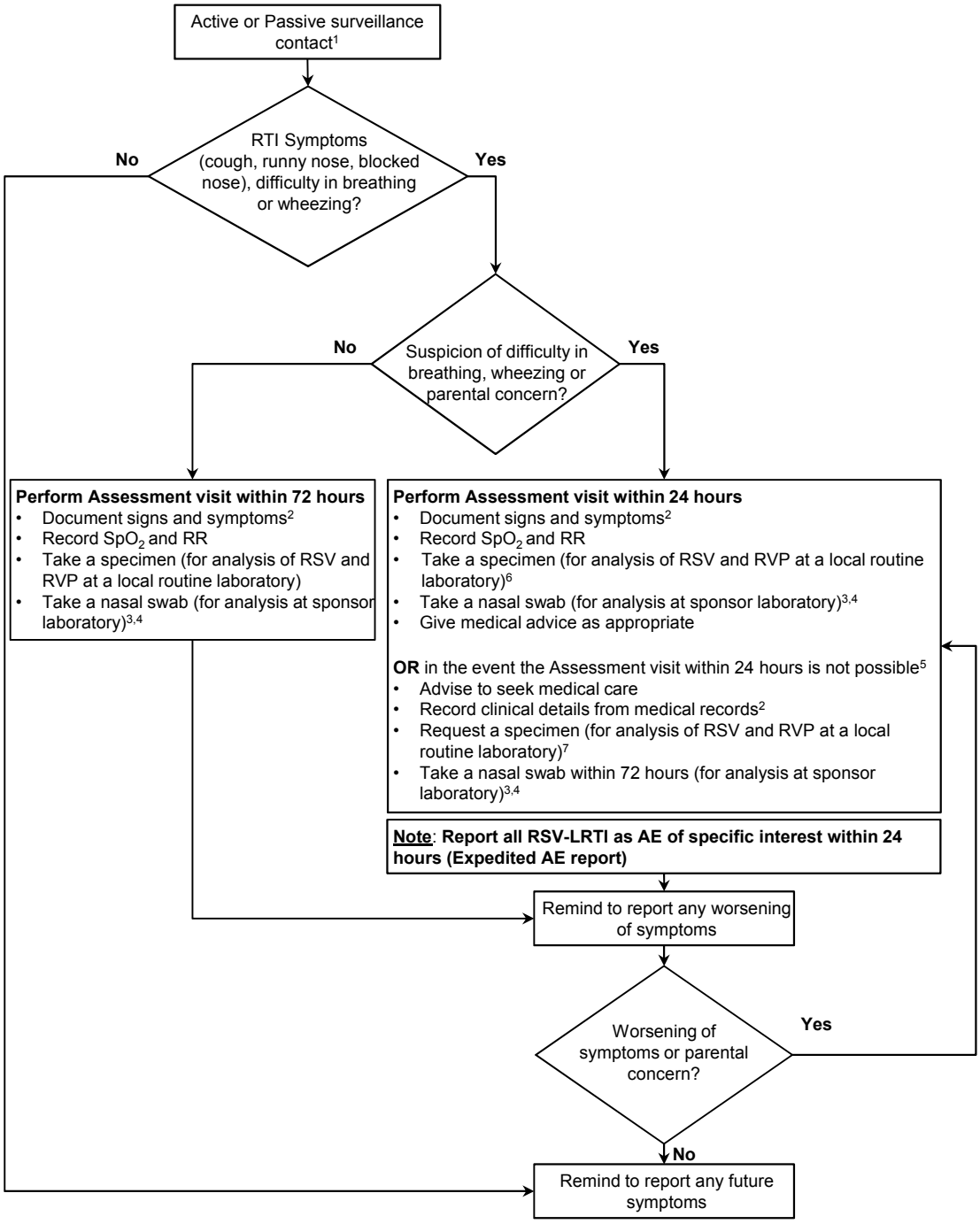
In addition to report these cases as AE of specific interest, the investigator should investigate the case according to the procedures for the surveillance of RSV-RTI (refer to Section 9.2) and document it in the eCRF. Of note a nasal swab should be taken for confirmatory testing of RSV infection by the sponsor.

9.2. Surveillance for RSV-RTI, difficulty in breathing and wheezing episodes

Each subject will be under surveillance for RSV-RTI, difficulty in breathing and wheezing episodes from the administration of Dose 1 (Visit 1) and will continue until the final visit (Visit 11). Surveillance will be performed via phone or e-mail contacts and assessment visits (refer to Figure 3). The passive and active surveillance contacts can also be made by/with the person designated by the parent(s)/LAR(s) (e.g. grandparents, nanny) as long as the parent(s)/LAR(s) have given approval.

The safety of the infant is paramount and for any reported illness, the investigator/study staff should assess the need for any intervention and provide this as part of regular healthcare or instruct/advise the parent(s)/LAR(s) where to obtain this care.

Figure 3 Decision tree for passive and active surveillance contacts



[1] Active contacts will take place weekly during the RSV season and every month outside the RSV season. Passive phone contacts from the carers to the investigator will take place when symptoms occur.
[2] Refer to the SPM for signs and symptoms to be collected.
[3] RSV-A/B quantitative PCR on all specimens.
[4] RVP (Multiplex PCR) on all specimens RSV-A/B positive and on all cases of confirmed LRTI according to case definition presented in Table 4.
[5] For example, in the event that the subject requires urgent medical evaluation and care or has travelled to another location,…
[6] In case of worsening of symptoms, optional sample if the previous specimen was RSV-positive.
[7] Where reasonably possible.

9.2.1. Passive surveillance

All subjects' parent(s)/LAR(s) will be instructed to contact investigator/study staff in case of any new RTI symptoms (cough, runny nose or blocked nose) or in case of any new difficulty in breathing or wheezing. They will be also reminded to record the start date and the end date of RTI symptoms on the RTI episode card.

- **If there is** any new difficulty in breathing, wheezing or parental concern, the investigator/study staff will schedule an assessment visit as soon as possible (refer to Section 9.2.4), but no later than 24 hours after the phone contact to ensure prompt assessment of the need for medical care.
- **If there is no suspicion** of difficulty in breathing, nor wheezing, nor parental concern, but there are any new symptoms of an RTI (cough, runny nose or blocked nose), the investigator/study staff will schedule an assessment visit as soon as possible (refer to Section 9.2.4), but no later than 72 hours after the phone contact. Performing the visit as soon as possible is important to assess the need for medical care, to ensure that early symptoms are captured, and also to collect an optimal nasal swab for the detection of RSV.
- All passive surveillance contacts will be document in the eCRF.
- In the event that it is not possible to schedule an assessment visit, which may arise if the family have travelled, then the assessment visit page of the eCRF should be filled in as completely as possible using available medical records.

9.2.2. Surveillance for asymptomatic RSV-RTI

- In order to detect asymptomatic RSV-RTI, monthly nasal swabs for analysis at sponsor laboratory will be performed for all subjects during the RSV season. This nasal swab will be tested by quantitative PCR.
- These nasal swabs may be performed in the subject's home or the investigators clinical facility as appropriate to the circumstances in the judgment of the investigator.
- All surveillance for asymptomatic RSV-RTI visits will be documented in the eCRF.

9.2.3. Active surveillance

There will be contact between the investigator/study staff and subject's parent(s)/LAR(s) on a regular basis (weekly during the RSV season and every month outside the RSV season). If there has not been a contact through a clinic visit/home visit/phone call (i.e. Visits 1, 2, 3, 4, 5, 6, 7, 8, 9, 10, 11), a passive surveillance contact (refer to Section 9.2.1), an assessment visit (refer to Section 9.2.4) or a surveillance for asymptomatic RSV infection (refer to Section 9.2.2), then the investigator/study staff will contact the subject's parent(s)/LAR(s). The active surveillance will be performed by phone, mobile phone or e-mail. During each active follow-up contact, the investigator/study staff will:

- Confirm with the subjects' parent(s)/LAR(s), if the subject has developed new RTI symptoms (cough, runny nose or blocked nose) and if he/she has developed any symptoms of difficulty in breathing or wheezing (during and between contacts).
  - **If there is** any new or ongoing difficulty in breathing, wheezing or parental concern, the investigator/study staff will schedule an assessment visit as soon as possible (refer to Section 9.2.4), but no later than 24 hours after the contact to ensure prompt assessment of the need for medical care. The investigator/study staff will remind the subjects' parent(s)/LAR(s) to record the start date and the end date of the RTI symptoms on the RTI episode card.
  - **If there is no suspicion** of difficulty in breathing, nor wheezing, nor parental concern but any new symptoms of an RTI (cough, runny nose or blocked nose) are present, the investigator/study staff will schedule an assessment visit as soon as possible (refer to Section 9.2.4), but no later than 72 hours after the contact. Performing the visit as soon as possible is important to assess the need for medical care, to ensure that early symptoms are captured, and also to collect a nasal swab for the detection of RSV.
- All active surveillance contacts will be document in the eCRF.
- In the event that it is not possible to schedule an assessment visit, which may arise if the family have travelled, then the assessment visit page of the eCRF should be filled in as completely as possible using available medical records.

9.2.4. Assessment visit

The purpose of the assessment visit is to objectively document signs and symptoms by an appropriately qualified person (i.e. medical or nursing), provide medical advice and to take a nasal swab for detection of RSV infection.

- Assessment visits may take place in the subject's home, the investigators clinical facility or a medical facility as appropriate to the circumstances in the judgment of the investigator.
- If the reported symptoms are already of a level of severity that urgent care is indicated, the parent(s)/LAR(s) should be redirected to the proper location to receive this care (e.g. Emergency Room) and an assessment visit could be scheduled to take place there at a suitable time.
- During the assessment visit, the investigator/study staff will evaluate the clinical signs and symptoms of the RTI. Signs and symptoms to be recorded in the eCRF are listed in the SPM.
- Specimens for testing of RSV and RVP at a local routine laboratory will be collected for all the subjects who have symptoms of an RTI.
- Nasal swabs for analysis at sponsor laboratory will be collected for all the subjects who have symptoms of an RTI. Note: all subjects experiencing an RSV-LRTI must be reported as an AE of specific interest in an expedited manner (refer to Section 9.4).

- Parent(s)/LAR(s) will be instructed to contact the study staff if the severity of the already existing symptoms increases or if the subject develops difficulty in breathing or wheezing and this may lead to a repeat assessment visit upon the judgment of the investigator.
- During subsequent surveillance contacts after an assessment visit, the status and evolution of the case will be followed until case resolution.
- Each assessment visit will be encoded in the eCRF.

9.2.5. Subject hospitalized for LRTI

Subjects admitted to hospital and treated for an LRTI will be documented fully as SAEs. In addition to establish the severity of the illness, the eCRF screen corresponding to the assessment visit will be completed as fully as possible from medical records. Signs and symptoms to be recorded in the eCRF are listed in the SPM. Study staff will collect a nasal swab for all the subjects who have been hospitalized with a RTI for analysis at sponsor laboratory. All subjects with clinical suspicion of LRTI must be tested for RSV and RVP using locally available test. Note: all subjects experiencing an RSV-LRTI must be reported as an AE of specific interest in an expedited manner (refer to Section 9.4).

Study staff will also collect a blood sample to be stored at sponsor's laboratory for future assessment of mechanism of illness (potential ERD). This will be done only for RSV-positive subjects using a locally available RSV test.

9.3. Detecting and recording adverse events, adverse events of specific interest and serious adverse events

9.3.1. Time period for detecting and recording adverse events, adverse events of specific interest and serious adverse events

All AEs starting within 30 days following administration of each dose of study vaccine (Day 0 to Day 29) must be recorded into the appropriate section of the eCRF, irrespective of intensity or whether or not they are considered vaccination-related.

The time period for collecting and recording SAEs will begin at the first receipt of study vaccine and will end at study conclusion. See Section 9.4 for instructions on reporting of SAEs.

All AEs/SAEs leading to withdrawal from the study will be collected and recorded from the time of the first receipt of study vaccines.

In addition to the above-mentioned reporting requirements and in order to fulfill international reporting obligations, SAEs that are related to study participation (i.e. protocol-mandated procedures, invasive tests, a change from existing therapy) or are related to a concurrent GSK medication/vaccine will be collected and recorded from the time the subject consents to participate in the study until she/he is discharged from the study.

The time period for collecting AEs of specific interest (RSV-LRTI) will begin at the first receipt of study vaccine and will end at study conclusion. The time period for collecting AEs of specific interest (spontaneous or excessive bleeding) will begin at the first receipt of study vaccine and will end 30 days after the last vaccination. See Section 9.4 for instructions on reporting of AEs of specific interest.

An overview of the protocol-required reporting periods for AEs, AEs of specific interest and SAEs is given in Table 18.

Table 18 Reporting periods for collecting safety information

Visit	SCR	V1	V2	V3		V4		V5	V6	V7		V8		V9	V10	V11
Days	-29 to 0	0	1	3	6	7	29	30	31	33	36	37	59	60	365	730
Solicited local and general AEs																
Unsolicited AEs																
Clinical examination with special attention given to the detection of episodes of spontaneous bleeding or easy bruising and evidence of bruising or petechiae*																
Examination of limb for petechiae induced by the tourniquet**																
AEs/SAEs leading to withdrawal from the study																
AE of specific interest (RSV-LRTI)																
AE of specific interest (spontaneous or excessive bleeding)																
SAEs																

Visit	SCR	V1	V2	V3		V4		V5	V6	V7		V8		V9	V10	V11
Days	-29 to 0	0	1	3	6	7	29	30	31	33	36	37	59	60	365	730
SAEs related to study participation (start at signature of informed consent form) or concurren GSK medication/vaccine																

SCR: Screening

* Clinical examination with special attention given to the detection of spontaneous bleeding or easy bruising and evidence of bruising or petechiae will be performed by the investigator/study staff on Day 1, Day 3, and Day 7 after each vaccination. The visits on Day 3 and Day 33 may be replaced by a phone call by the investigator/study staff to inquire of the infants' general health and to solicit reports of a rash, spontaneous bleeding or easy bruising. Subjects' parent(s)/LAR(s) will be instructed to contact the investigator/study staff if their child presents symptoms of spontaneous bleeding or easy bruising or if their child develops a rash in the month following vaccination, in order to detect any thrombocytopenic petechiae or purpura.

**On Day 7 and/or Day 37, special attention to detection of petechiae or evidence of petechiae induced by the tourniquet will be given in case hematology testing will be performed on that day.

9.3.2. Post-Study adverse events and serious adverse events

A post-study AE/SAE is defined as any event that occurs outside of the AE/SAE reporting period defined in Table 18. Investigators are not obligated to actively seek AEs or SAEs in former study participants. However, if the investigator learns of any SAE at any time after a subject has been discharged from the study, and he/she considers the event reasonably related to the investigational vaccines, the investigator will promptly notify the Study Contact for Reporting SAEs.

9.3.3. Evaluation of adverse events and serious adverse events

9.3.3.1. Active questioning to detect adverse events and serious adverse events

As a consistent method of collecting AEs, the subject's parent(s)/LAR(s) should be asked a non-leading question such as:

'*Has your child acted differently or felt different in any way since receiving the vaccines or since the last visit?*'

When an AE/SAE occurs, it is the responsibility of the investigator to review all documentation (e.g. hospital progress notes, laboratory and diagnostics reports) relative to the event. The investigator will then record all relevant information regarding an AE/SAE in the eCRF. The investigator is not allowed to send photocopies of the subject's medical records to GSK Biologicals instead of appropriately completing the eCRF. However, there may be instances when copies of medical records for certain cases are requested by GSK Biologicals. In this instance, all subject identifiers will be blinded on the copies of the medical records prior to submission to GSK Biologicals.

The investigator will attempt to establish a diagnosis pertaining to the event based on signs, symptoms, and/or other clinical information. In such cases, the diagnosis should be documented as the AE/SAE and not the individual signs/symptoms.

9.3.3.2. Assessment of adverse events

9.3.3.2.1. Assessment of intensity

The intensity of the following solicited AEs will be assessed as described:

Table 19 Intensity scales for solicited symptoms in infants

Infants		
Adverse Event	**Intensity grade**	**Parameter**
Pain at injection site	0	None
	1	Mild: Minor reaction to touch
	2	Moderate: Cries/protests on touch
	3	Severe: Cries when limb is moved/spontaneously painful
Redness at injection site		Record greatest surface diameter in mm
Swelling at injection site		Record greatest surface diameter in mm
Fever*		Record temperature in °C/°F
Irritability/Fussiness	0	Behavior as usual
	1	Mild: Crying more than usual/no effect on normal activity
	2	Moderate: Crying more than usual/interferes with normal activity
	3	Severe: Crying that cannot be comforted/prevents normal activity
Drowsiness	0	Behavior as usual
	1	Mild: Drowsiness easily tolerated
	2	Moderate: Drowsiness that interferes with normal activity
	3	Severe: Drowsiness that prevents normal activity
Loss of appetite	0	Appetite as usual
	1	Mild: Eating less than usual/no effect on normal activity
	2	Moderate: Eating less than usual/interferes with normal activity
	3	Severe: Not eating at all

*Fever is defined as temperature ≥ 37.5°C / 99.5°F for oral, axillary or tympanic route, or ≥ 38.0°C / 100.4°F for rectal route. The preferred route for recording temperature in this study will be axillary.

The maximum intensity of local injection site redness and swelling will be scored at GSK Biologicals as follows:

0 : None
1 : < 5 mm
2 : 5 to 20 mm
3 : > 20 mm

The maximum intensity of fever will be scored at GSK Biologicals as follows:

0 : < 37.5°C/99.5°F
1 : ≥ 37.5°C/99.5°F to ≤ 38.5°C/101.3°F
2 : > 38.5°C/101.3°F to ≤ 39.5°C/103.1°F
3 : > 39.5°C/103.1°F

The investigator will assess the maximum intensity that occurred over the duration of the event for all unsolicited AEs (including SAEs) recorded during the study. The assessment will be based on the investigator's clinical judgment.

The intensity should be assigned to one of the following categories:

1 (mild)	=	An AE which is easily tolerated by the subject, causing minimal discomfort and not interfering with everyday activities.
2 (moderate)	=	An AE which is sufficiently discomforting to interfere with normal everyday activities.
3 (severe)	=	An AE which prevents normal, everyday activities. Such an AE would, for example, prevent attendance at kindergarten/a day-care center and would cause the parent(s)/LAR(s) to seek medical advice.

An AE that is assessed as Grade 3 (severe) should not be confused with a SAE. Grade 3 is a category used for rating the intensity of an event; and both AEs and SAEs can be assessed as Grade 3. An event is defined as 'serious' when it meets one of the pre-defined outcomes as described in Section 9.1.2.

9.3.3.2.2. Assessment of causality

The investigator is obligated to assess the relationship between investigational vaccine and the occurrence of each AE/SAE. The investigator will use clinical judgment to determine the relationship. Alternative plausible causes, such as natural history of the underlying diseases, concomitant therapy, other risk factors, and the temporal relationship of the event to the investigational vaccines will be considered and investigated. The investigator will also consult the IB to determine his/her assessment.

There may be situations when a SAE has occurred and the investigator has minimal information to include in the initial report to GSK Biologicals. However, it is very important that the investigator always makes an assessment of causality for every event prior to submission of the Expedited Adverse Events Report to GSK Biologicals. The investigator may change his/her opinion of causality in light of follow-up information and update the SAE information accordingly. The causality assessment is one of the criteria used when determining regulatory reporting requirements.

In case of concomitant administration of multiple vaccines/products, it may not be possible to determine the causal relationship of general AEs to the individual vaccine administered. The investigator should, therefore, assess whether the AE could be causally related to vaccination rather than to the individual vaccines.

All solicited local (injection site) reactions will be considered causally related to vaccination. Causality of all other AEs should be assessed by the investigator using the following question:

Is there a reasonable possibility that the AE may have been caused by the investigational vaccine?

YES	:	There is a reasonable possibility that the vaccine contributed to the AE.
NO	:	There is no reasonable possibility that the AE is causally related to the administration of the study vaccine. There are other, more likely causes and administration of the study vaccine is not suspected to have contributed to the AE.

If an event meets the criteria to be determined as 'serious' (see Section 9.1.2), additional examinations/tests will be performed by the investigator in order to determine ALL possible contributing factors for each SAE.

Possible contributing factors include:

- Medical history.
- Other medication.
- Protocol required procedure.
- Other procedure not required by the protocol.
- Lack of efficacy of the vaccines, if applicable.
- Erroneous administration.
- Other cause (specify).

9.3.3.3. Assessment of outcomes

The investigator will assess the outcome of all unsolicited AEs (including SAEs) recorded during the study as:

- Recovered/resolved.
- Recovering/resolving.
- Not recovered/not resolved.
- Recovered with sequelae/resolved with sequelae.
- Fatal (SAEs only).

9.3.3.4. Medically attended visits

For each solicited and unsolicited symptom the subject experiences, the subject's parent(s)/LAR(s) will be asked if the subject received medical attention defined as hospitalization, or an otherwise unscheduled visit to or from medical personnel for any reason, including emergency room visits. This information will be recorded in the eCRF.

9.4. Reporting of serious adverse events and adverse events of specific interest

9.4.1. Prompt reporting of serious adverse events and adverse events of specific interest to GSK Biologicals

SAEs that occur in the time period defined in Section 9.3 will be reported promptly to GSK within the timeframes described in Table 20, once the investigator determines that the event meets the protocol definition of a SAE.

AEs of specific interest (RSV-LRTI and spontaneous or excessive bleeding) that occur in the time period defined in Section 9.3 will be reported promptly to GSK within the timeframes described in Table 20.

Table 20 Timeframes for submitting serious adverse event and adverse events of specific interest reports to GSK Biologicals

Type of Event	Initial Reports		Follow-up of Relevant Information on a Previous Report	
	Timeframe	Documents	Timeframe	Documents
SAEs	24 hours*‡	electronic Expedited Adverse Events Report	24 hours*	electronic Expedited Adverse Events Report
AEs of specific interest	24 hours*‡	electronic Expedited Adverse Events Report	24 hours*	electronic Expedited Adverse Events Report

* Timeframe allowed after receipt or awareness of the information.
‡ The investigator will be required to confirm review of the SAE/AEs of specific interest causality by ticking the 'reviewed' box in the electronic Expedited Adverse Events Report within 72 hours of submission of the SAE/AEs of specific interest.

9.4.2. Contact information for reporting serious adverse events and adverse events of specific interest

Study Contact for Reporting SAEs and AEs of specific interest
Refer to the local study contact information document.
Back-up Study Contact for Reporting SAEs and AEs of specific interest
24/24 hour and 7/7 day availability: **GSK Biologicals Clinical Safety & Pharmacovigilance** Outside US & Canada sites: Fax: PPD or PPD Email address: PPD US sites only: Fax: PPD Canadian sites only: Fax: PPD

9.4.3. Reporting of adverse events of specific interest (RSV-LRTI and spontaneous or excessive bleeding) to GSK Biologicals

Once an Investigator becomes aware that an AE of specific interest (RSV-LRTI and spontaneous or excessive bleeding) has occurred in a subject, the Investigator (or designate) must complete the information in the electronic Expedited Adverse Events Report WITHIN 24 HOURS. The report will always be completed as thoroughly as possible with all available details of the event. Even if the Investigator does not have all information regarding an AE of specific interest (RSV-LRTI and spontaneous or excessive bleeding), the report should still be completed within 24 hours. Once additional relevant information is received, the report should be updated WITHIN 24 HOURS.

The Investigator will always provide an assessment of causality at the time of the initial report. The Investigator will be required to confirm the review of causality by ticking the 'reviewed' box in the electronic Expedited Adverse Events Report within 72 hours of submission of the AE of specific interest.

Refer to Section 9.4.4.1 for the back-up system in case the electronic reporting system does not work.

9.4.4. Completion and transmission of SAE reports to GSK Biologicals

Once an investigator becomes aware that a SAE has occurred in a study subject, the investigator (or designate) must complete the information in the electronic Expedited Adverse Events Report WITHIN 24 HOURS. The report will always be completed as thoroughly as possible with all available details of the event. Even if the investigator does not have all information regarding a SAE, the report should still be completed within 24 hours. Once additional relevant information is received, the report should be updated WITHIN 24 HOURS.

The investigator will always provide an assessment of causality at the time of the initial report. The investigator will be required to confirm the review of the SAE causality by ticking the 'reviewed' box in the electronic Expedited Adverse Events Report within 72 hours of submission of the SAE.

9.4.4.1. Back-up system in case the electronic reporting system does not work

If the electronic reporting system does not work, the investigator (or designate) must complete, then date and sign a paper Expedited Adverse Events Report and fax it to the Study Contact for Reporting SAEs (refer to the Sponsor Information) or to GSK Biologicals Clinical Safety and Pharmacovigilance department within 24 hours.

This back-up system should only be used if the electronic reporting system is not working and NOT if the system is slow. As soon as the electronic reporting system is working again, the investigator (or designate) must complete the electronic Expedited Adverse Events Report within 24 hours. The final valid information for regulatory reporting will be the information reported through the electronic SAE reporting system.

9.4.5. Updating of serious adverse event and adverse event of specific interest information after removal of write access to the subject's eCRF

When additional SAE or AEs of specific interest information is received after removal of the write access to the subject's eCRF, new or updated information should be recorded on the appropriate paper report, with all changes signed and dated by the investigator. The updated report should be faxed to the Study Contact for Reporting SAEs (refer to the Sponsor Information) or to GSK Biologicals Clinical Safety and Pharmacovigilance department within the designated reporting time frames specified in Table 20.

9.4.6. Regulatory reporting requirements for serious adverse events

The investigator will promptly report all SAEs to GSK in accordance with the procedures detailed in Section 9.4.1. GSK Biologicals has a legal responsibility to promptly notify, as appropriate, both the local regulatory authority and other regulatory agencies about the safety of a product under clinical investigation. Prompt notification of SAEs by the investigator to the Study Contact for Reporting SAEs is essential so that legal obligations and ethical responsibilities towards the safety of other subjects are met.

Investigator safety reports are prepared according to the current GSK policy and are forwarded to investigators as necessary. An investigator safety report is prepared for a SAE(s) that is both attributable to the investigational vaccines and unexpected. The purpose of the report is to fulfill specific regulatory and GCP requirements, regarding the product under investigation.

9.5. Follow-up of adverse events, serious adverse events and adverse events of specific interest

9.5.1. Follow-up during the study

After the initial AE, AE of specific interest or SAE report, the investigator is required to proactively follow each subject and provide additional relevant information on the subject's condition to GSK Biologicals (within 24 hours for AE of specific interest and SAEs; refer to Table 20).

All AE of specific interest (RSV-LRTI) and SAEs documented at a previous visit/contact and designated as not recovered/not resolved or recovering/resolving will be reviewed at subsequent visits/contacts until the end of the study.

All AE of specific interest (spontaneous or excessive bleeding) documented at a previous visit/contact and designated as not recovered/not resolved or recovering/resolving will be reviewed at subsequent visits/contacts until 30 days after the last vaccination.

All AEs documented at a previous visit/contact and designated as not recovered/not resolved or recovering/resolving will be reviewed at subsequent visits/contacts until 30 days after the last vaccination.

9.5.2. Follow-up after the subject is discharged from the study

The investigator will follow subjects:

- with SAEs, with AEs of specific interest or subjects withdrawn from the study as a result of an AE, until the event has resolved, subsided, stabilized, disappeared, or until the event is otherwise explained, or the subject is lost to follow-up.

If the investigator receives additional relevant information on a previously reported SAE, he/she will provide this information to GSK Biologicals using electronic Expedited Adverse Events Report.

GSK Biologicals may request that the investigator performs or arranges the conduct of additional clinical examinations/tests and/or evaluations to elucidate as fully as possible the nature and/or causality of the AE or SAE. The investigator is obliged to assist. If a subject dies during participation in the study or during a recognized follow-up period, GSK Biologicals will be provided with any available post-mortem findings, including histopathology.

9.6. Treatment of adverse events

Treatment of any AE is at the sole discretion of the investigator and according to current good medical practice. Any medication administered for the treatment of an AE should be recorded in the subject's eCRF (refer to Section 7.6).

9.7. Unblinding

GSK Biologicals' policy (which incorporates ICH E2A guidance, EU Clinical Trial Directive and US Federal Regulations) is to unblind the report of any SAE which is unexpected and attributable/suspected to be attributable to the investigational vaccines, prior to regulatory reporting. The GSK Biologicals' Central Safety Physician is responsible for unblinding the treatment assignment in accordance with the specified timeframes for expedited reporting of SAEs (refer to Section 9.4.1).

9.8. Emergency unblinding

Unblinding of a subject's individual treatment code should occur only in the case of a medical emergency, or in the event of a serious medical condition, when knowledge of the treatment is essential for the clinical management or welfare of the subject, as judged by the investigator.

The emergency unblinding process consists of the automated system SBIR that allows the investigator to have unrestricted, immediate and direct access to the subject's individual study treatment.

The investigator has the option of contacting a GSK Biologicals' On-call Central Safety Physician (or Backup) if he/she needs medical advice or needs the support of GSK to perform the unblinding (i.e. he/she cannot access SBIR).

Any emergency unblinding must be fully documented by using the Emergency Unblinding Documentation Form, which must be appropriately completed by the investigator and sent within 24 hours to GSK Biologicals.

GSK Biologicals' Contact information for Emergency Unblinding **24/24 hour and 7/7 day availability**
GSK Biologicals' Central Safety Physician: Outside US: PPD (GSK Biologicals Central Safety Physician on-call) For US: PPD (GSK Biologicals Central Safety Physician on-call) **GSK Biologicals' Central Safety Physician Back-up:** Outside US: PPD US only: PPD **Emergency Unblinding Documentation Form transmission:** Outside US: Fax: PPD or PPD US only: Fax: PPD

9.9. Subject card

Study subjects' parent(s)/LAR(s) must be provided with the address and telephone number of the main contact for information about the clinical study.

The investigator (or designate) must therefore provide a "subject card" to each subject's parent(s)/LAR(s). In an emergency situation this card serves to inform the responsible attending physician that the subject is in a clinical study and that relevant information may be obtained by contacting the investigator.

Subjects' parent(s)/LAR(s) must be instructed to keep subject cards in their possession at all times.

9.10. Holding rules and safety monitoring

The investigational ChAd155-RSV vaccine has never been administered to infants. Safety precautions such as staggered enrolment, dose-escalation, limited vaccination, safety evaluations by an iSRC and an IDMC, and study holding rules have therefore been defined.

9.10.1. Limited vaccination

Limited vaccination in Steps 1, 2 and 3:

- During vaccination days, vaccination will be limited to maximum 10 infants per day.

In the same investigational center, all infants should be vaccinated sequentially and at least 60 minutes apart to allow monitoring of any acute events (e.g. hypersensitivity reaction). All infants should be closely observed (visual follow-up as well as measurement of vital signs) for at least 60 minutes after vaccination. Vital signs are body temperature, HR, and RR. Vital signs are preferably measured when the infant is calm. In addition, to monitor delayed potential reactions following vaccination, there will be a 48-hour observation period following Dose 1 and Dose 2 administration to the first eight infants (corresponding to three to five ChAd155-RSV vaccine recipients) in each Step (1, 2, and 3) before continuing vaccination of more infants (i.e. from the ninth infant). From the ninth infant, infants will be vaccinated sequentially, separated by a minimum interval of 60 minutes. Refer to the SPM for more details about the 48-hour observation period after the first eight subjects vaccinated in each Step.

9.10.2. Internal safety review committee (iSRC) oversight

This study will be overseen by an iSRC operating under a charter. Core members of the iSRC will include a GSK Biologicals' safety physician, a CRDL, and a biostatistician who are not otherwise involved in the conduct of the project. The iSRC safety reviews will be conducted using unblinded data. The iSRC has access to the subject randomization and reviews unblinded data.

The iSRC will review all accumulating safety data three weeks after the start of vaccination and then about every three weeks (until the IDMC has reviewed all safety data up to 30 days after administration of Dose 2 in Step 3 (i.e. Day 60). The iSRC members will determine if a safety signal should be escalated to the IDMC. The iSRC members will determine whether any of the predefined holding rules are met (refer to Section 9.10.5) or if there is any other safety signal. In this case, vaccination in the study will be immediately put on hold (refer to Section 9.10.6).

The iSRC will receive the following safety data within 48 hours upon GSK becoming aware of:

- Fatal SAEs.
- Life-threatening SAEs.

- Related SAEs.
- SAEs occurring within 30 days of vaccination.
- LRTI associated with RSV infection (AE of specific interest) occurring from Day 0 to Day 730.
- Spontaneous or excessive bleeding (AE of specific interest) occurring within 30 days after each vaccination.

In addition, the iSRC will receive:

- Unblinded summary reports of accumulated solicited AEs, unsolicited AEs, results of laboratory monitoring of hematology and biochemistry.
- New information that may adversely affect the safety of the subjects or the conduct of the study.
- All subsequent protocol amendments, informed consent changes or revisions or other documents originally submitted for review.
- All subsequent protocol administrative changes (for information).

9.10.3. Independent Data Monitoring Committee (IDMC) oversight

This study will be overseen by an IDMC operating under a charter. Overall, the role of the IDMC includes the review and protection of data integrity and rights and safety of study participants throughout the study period. It will provide initial, regular, and closing advice to GSK Biologicals on medical, ethical, scientific and safety-related issues. Its advice will be based on the interpretation of study data with reference to the study protocol.

The IDMC will review the protocol and statistical analysis plan. Meetings will be documented and minutes of open sessions of the IDMC meetings made available to the sponsor. The IDMC may, if deemed necessary, convene a meeting with, or request further information from the principal investigators and GSK Biologicals' designated project representatives at any stage of the study.

The IDMC may recommend to the sponsor to suspend the enrolment to the study and/or vaccination based on their review of safety data arising in this study (refer to Section 9.10.6).

The IDMC safety reviews will be conducted using unblinded data. The IDMC will review all available safety data while taking into account any other findings that could have an impact on the safety of the subjects.

The IDMC members will determine whether any of the predefined study holding rules are met (refer to Section 9.10.5) or if there is any other safety signal. If this is the case, vaccination in the study will be immediately put on hold. If no safety signal is observed, the favorable outcome of the safety evaluation authorizing the investigator to proceed with vaccination of infants as outlined in Figure 1, will be also documented and provided in writing.

- The IDMC will receive the following safety data within 48 hours upon GSK becoming aware of:
  - Fatal SAEs occurring from Day 0 to Day 730.
  - Life-threatening SAEs occurring from Day 0 to Day 730.
  - Related SAEs occurring from Day 0 to Day 730.
  - SAEs occurring within 30 days of vaccination.
  - LRTI associated with RSV infection (AE of specific interest) occurring from Day 0 to Day 730.
  - Spontaneous or excessive bleeding (AE of specific interest) occurring within 30 days after each vaccination.
- The IDMC will receive the following safety data monthly throughout the period of vaccination (up to 30 days after administration of Dose 2 in Step 3, i.e. Day 60):
  - Summary reports of solicited and unsolicited AEs, SAEs and laboratory toxicity.
- The IDMC will receive the following safety data approximately monthly from beginning of the first RSV season and the frequency will be adapted based on the instruction of the IDMC:
  - Cumulative reports of the incidence of RSV-RTI, RSV-LRTI, RSV-severe LRTI and RSV-RTI leading to hospitalization.
  - Cumulative tables of incidence of all SAEs.

In addition, the IDMC will receive from GSK Biologicals:

- New information that may adversely affect the safety of the subjects or the conduct of the study.
- All subsequent protocol amendments, informed consent changes or revisions or other documents originally submitted for review.
- All subsequent protocol administrative changes (for information).

9.10.4. Dose-escalation and safety evaluations by iSRC and IDMC

The study will be conducted in a staggered manner to ensure maximum safety of the participating infants progressing sequentially through the 3 dose levels (Figure 1):

- **Step 1**: vaccination of ***a maximum of*** 32 infants (approximately 16 infants in the RSV-Ld group and approximately 16 infants in the placebo-Ld group). An iSRC will review all accumulating safety data three weeks after the start of vaccination in Step 1 and then about every three weeks (until the IDMC has reviewed all safety data up to 30 days after administration of Dose 2 in Step 3 (i.e. Day 60). The IDMC will review all accumulating reactogenicity and safety data monthly throughout the period of vaccination (up to 30 days after administration of Dose 2 in Step 3, i.e. Day 60). Dose escalation ***to Step 2*** will proceed after ***completion of vaccination f***or 32 subjects, and in the absence of a ***significant*** safety concern detected by the IDMC in

the regular monitoring of accumulating safety data. ***However, in the absence of a significant safety concern detected in the regular monitoring of accumulating safety data on at least 16 subjects, the IDMC may recommend for dose escalation to proceed without the requirement of enrolling and evaluating the full group size of 32 subjects. A subsequent dose escalation will proceed after administration of two doses of the vaccine to between a maximum of 32 to a minimum of 16 subjects in the absence of a significant safety concern detected by the IDMC in the regular monitoring of accumulating safety data.*** It will be ensured that, as a minimum the IDMC will have reviewed safety data of at least 16 subjects for two doses (in Step 1) and given their formal approval prior to enrolling subjects to the next dose level (Step 2). **(Amended 10 December 2017)**

- **Step 2**: vaccination of ***a maximum of*** 32 infants (approximately 16 infants in the RSV-Md group and approximately 16 infants in the placebo-Md group). An iSRC will review all accumulating safety data three weeks after the start of vaccination in Step 2 and then about every three weeks (until the IDMC has reviewed all safety data up to 30 days after administration of Dose 2 in Step 3 (i.e. Day 60). The IDMC will review accumulating reactogenicity and safety data monthly throughout the period of vaccination (up to 30 days after administration of Dose 2 in Step 3, i.e. Day 60). Dose escalation ***to Step 3*** will proceed after administration ***of two doses*** of the vaccine to 32 subjects, and in the absence of a ***significant*** safety concern detected by the IDMC in the regular monitoring of accumulating safety data. ***However, as for the dose escalation from Step 1 to Step 2, in the absence of a significant safety concern detected in the regular monitoring of accumulating safety data on at least 16 subjects, the IDMC may again allow for dose escalation to proceed without the requirement of enrolling and evaluating the full group size of 32 subjects. A subsequent dose escalation will proceed after administration of two doses of the vaccine to between a maximum of 32 to a minimum of 16 subjects in the absence of a significant safety concern detected by the IDMC in the regular monitoring of accumulating safety data.*** It will be ensured that, as a minimum the IDMC will have reviewed safety data of at least 16 subjects for two doses (in Step 2) and given their formal approval prior to enrolling subjects to the next dose level (Step 3). **(Amended 10 December 2017)**
- **Step 3**: vaccination of 32 infants (approximately 16 infants in the RSV-Hd group and approximately 16 infants in the placebo-Hd group). An iSRC will review all accumulating safety data three weeks after the start of vaccination in Step 3 and then about every three weeks (until the IDMC has reviewed all safety data up to 30 days after administration of Dose 2 in Step 3 (i.e. Day 60). A review of the entire set of safety data from Steps 1, 2 and 3 will be performed by the IDMC after availability of safety data up to 30 days after administration of Dose 2 in Step 3 (i.e. Day 60).

Refer to Section 9.10.2 for iSRC oversight and to Section 9.10.3 for IDMC oversight.

9.10.5. Holding rules

The safety holding rules which will be assessed by the investigator are defined in Table 21 and the safety holding rules which will be assessed by the iSRC and the IDMC during the safety evaluation are defined in Table 22.

Table 21 Holding rules assessed by the investigator

Holding Rule	Event	Number of infants/group		
		Step 1	Step 2	Step 3
1a	Death or any life-threatening serious adverse event (SAE) that can be causally related to vaccination, according to investigator's assessment.	≥ 1	≥ 1	≥ 1
1b	Any withdrawal from the study (by investigator or parent(s)/LAR(s) of the subject request) following a Grade 3 AE that cannot reasonably be attributed to a cause other than vaccination.	≥ 1	≥ 1	≥ 1
1c	Any local or general solicited AE leading to hospitalization that cannot reasonably be attributed to a cause other than vaccination.	≥ 1	≥ 1	≥ 1
1d	Within 30 days post-vaccination, Any spontaneous local or general bleeding AND Thrombocytopenia < 50000/mm^3	≥ 1	≥ 1	≥ 1

If an investigator detects one of the holding rules mentioned above, he/she will immediately put the enrolment or the vaccination on hold (refer to Section 9.10.6) and he/she will immediately inform the sponsor and enter the data in the eCRF. It is sponsor's responsibility to put the enrolment or the vaccination on hold at all sites.

Table 22 Holding rules during the planned iSRC and IDMC evaluation

Holding Rule	Event	Number of infants/group		
		Step 1	Step 2	Step 3
2a	Any Grade 3 solicited local AE lasting 48 hours or more in an investigational RSV vaccine group, within the 7-day (Days 0-6) post-vaccination period.	≥ 25% & ≥ 2 infants/group	≥ 25% & ≥ 2 infants/group	≥ 25% & ≥ 2 infants/group
2b	Any Grade 3 solicited general AE lasting 48 hours or more in an investigational RSV vaccine group, that cannot reasonably be attributed to a cause other than vaccination, within the 7-day (Days 0-6) post-vaccination period.	≥ 25% & ≥ 2 infants/group	≥ 25% & ≥ 2 infants/group	≥ 25% & ≥ 2 infants/group
2c	Any ≥ Grade 3 unsolicited AE in an investigational RSV vaccine group, that cannot reasonably be attributed to a cause other than vaccination, within the 7-day (Days 0-6) post-vaccination period OR Any ≥ Grade 3 abnormality in pre-specified hematological or biochemical laboratory parameters in an investigational RSV vaccine group, if it cannot reasonably be attributed to a cause other than vaccination, up to the Day 7 post-vaccination visit[1]	≥ 25% & ≥ 2 infants/group	≥ 25% & ≥ 2 infants/group	≥ 25% & ≥ 2 infants/group

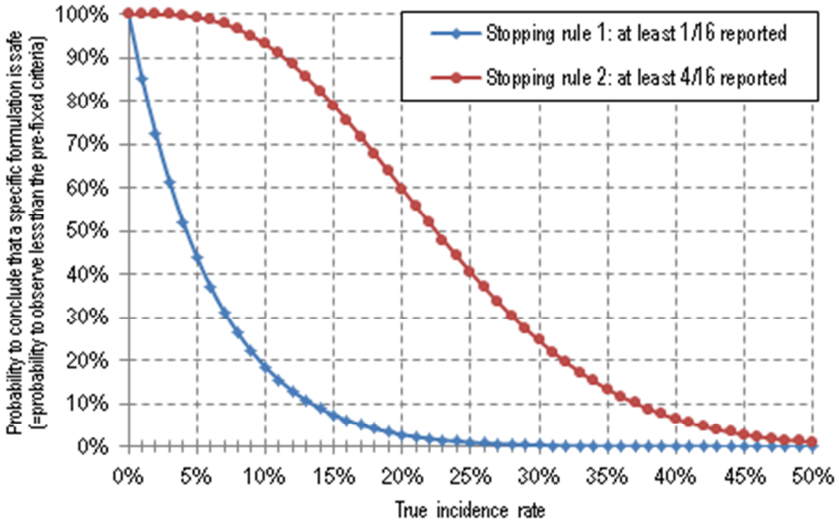
[1] Refer to Appendix C.

Of note, no formal holding rules will be applied for other safety data collected. However, these data will also be reviewed by the iSRC and the IDMC in order to allow for an overall assessment of the benefit/risk ratio of vaccination.

Risk assessment

Figure 4 gives the probability of not meeting holding rule 1-2 for 16 subjects per study group in each step.

Figure 4 Evaluations based on 16 subjects - Risk assessment curve for one formulation based on the proposed safety holding rules



The above figure illustrates that, with ***a maximum of*** 16 subjects per study group **(Amended 10 December 2017)**:

- Each holding rule 1 (a - d) has more than 85% chance of not being met for vaccination with a true incidence rate below 1% and has more than 80% chance of being met for vaccination with a true incidence rate above 10%.
- Each holding rule 2 (a - c) has more than 90% chance of not being met for vaccination with a true incidence rate below 10% and more than 60% chance of being met for vaccination with a true incidence rate above 25%.

9.10.6. Procedure if the trial is put on hold

If the trial is put on hold by the investigator, iSRC or IDMC because a pre-defined holding rule is met or because of a safety concern, then all enrolment in the study and all vaccination will cease immediately, but all other procedures relating to safety, immunology and disease monitoring will continue. The IDMC will review all available safety information and may ask for additional information to be provided by the investigators or the sponsor. The IDMC will make a recommendation to the sponsor whether the study should be stopped permanently, modified or continued unchanged.

The sponsor will review all data and IDMC recommendation and will decide whether to stop permanently, modify or continue the conduct of the study. The decision of the Vaccine safety monitoring board (VSMB) regarding the further conduct of the study will be documented and provided in writing to the investigators.

10. SUBJECT COMPLETION AND WITHDRAWAL

10.1. Subject completion

A subject who returns for the concluding visit foreseen in the protocol is considered to have completed the study.

10.2. Subject withdrawal

Withdrawals will not be replaced.

10.2.1. Subject withdrawal from the study

From an analysis perspective, a 'withdrawal' from the study refers to any subject who did not come back for the concluding visit foreseen in the protocol.

All data collected until the date of withdrawal/last contact of the subject will be used for the analysis.

A subject is considered a 'withdrawal' from the study when no study procedure has occurred, no follow-up has been performed and no further information has been collected for this subject from the date of withdrawal/last contact.

Investigators will make an attempt (e.g. three telephone calls and a certified letter to the last known address) to contact those subjects who do not return for scheduled visits or follow-up.

Information relative to the withdrawal will be documented in the eCRF. The investigator will document whether the decision to withdraw a subject from the study was made by the subject's parent(s)/LAR(s), or by the investigator, as well as which of the following possible reasons was responsible for withdrawal:

- SAE.
- Non-SAE.
- Protocol violation (specify).
- Consent withdrawal, not due to an AE*.
- Moved from the study area.
- Lost to follow-up.
- Other (specify).

*In case a subject is withdrawn from the study because the subject's parent(s)/LAR(s) has withdrawn consent, the investigator will document the reason for withdrawal of consent, if specified by the subject's parent(s)/LAR(s), in the eCRF.

Subjects who are withdrawn from the study because of SAEs/AEs must be clearly distinguished from subjects who are withdrawn for other reasons. Investigators will follow subjects who are withdrawn from the study as result of a SAE/AE until resolution of the event (see Section 9.5.2).

10.2.2. Subject withdrawal from investigational vaccine

A 'withdrawal' from the investigational vaccine refers to any subject who does not receive the complete treatment, i.e. when no further planned dose is administered from the date of withdrawal. A subject withdrawn from the investigational vaccine may not necessarily be withdrawn from the study as further study procedures or follow-up may be performed (safety or immunogenicity) if planned in the study protocol. The investigator should make all efforts to ensure that the subjects remain in the study to ensure a proper safety follow-up and medical care if needed.

Information relative to premature discontinuation of the investigational vaccine will be documented on the Vaccine Administration screen of the eCRF. The investigator will document whether the decision to discontinue further vaccination was made by the subject's parent(s)/LAR(s), or by the investigator, as well as which of the following possible reasons was responsible for withdrawal:

- SAE.
- Non-SAE.
- Other (specify).

10.3. Extension study

At the end of the study (study conclusion visit), the investigator will ask each subject's parent(s)/LAR(s) if they are interested to allow the subject to participate in a booster study/long-term study. If subject's parent(s)/LAR(s) are not interested in participating in the booster study/long-term study the reason for refusal will be documented in the subject's eCRF.

10.4. Screen and baseline failures

Screening failures are defined as subjects who are withdrawn from the study after giving informed consent, but who do not meet the inclusion and exclusion criteria.

The following information will be collected for screening failures:

- Informed consent.
- Inclusion/exclusion criteria.
- Demographic data.
- Medical history.

- Physical examination.
- Growth monitoring.
- Blood samples for RSV serostatus, hematology and biochemistry, CMI response and humoral response.
- SAEs related to study participation, to concomitant use of GSK products or any fatal SAEs.
- Screening conclusion.

11. STATISTICAL METHODS

11.1. Primary endpoint

- Occurrence of AEs from first vaccination (Day 0) up to Day 60.
  - Occurrence of each solicited local and general AE, during a 7-day follow-up period after each vaccination (i.e. the day of vaccination and 6 subsequent days).
  - Occurrence of any unsolicited AE, during a 30-day follow-up period after each vaccination (i.e. the day of vaccination and 29 subsequent days).
  - Occurrence of any SAE from Day 0 up to Day 60.
  - Occurrence of episode of spontaneous or excessive bleeding (AE of specific interest), during a 30-day follow-up period after each vaccination.
  - Occurrence of any hematological (hemoglobin level, white blood cells and platelets) laboratory abnormalities at Screening, Day 1, Day 7, Day 30, Day 31, Day 37, and Day 60.
  - Occurrence of any biochemical (alanine aminotransferase, aspartate aminotransferase and creatinine) laboratory abnormalities at Screening, Day 30, and Day 60.

11.2. Secondary endpoints

- Occurrence of SAEs from study start (Day 0) up to study conclusion (Day 730).
- Occurrence of RSV-LRTI (AE of specific interest) as from Dose 1 administration up to study conclusion (Day 730).
- Occurrence of RSV-RTI, RSV-LRTI, severe RSV-LRTI (according to standardized case definitions) as from Dose 1 administration up to study conclusion (Day 730).
- Magnitude of the CMI response to the investigational RSV vaccine, pre-vaccination (Screening), post-Dose 1 (Day 30) and post-Dose 2 (Day 60 and Day 365).
  - CD3+/CD4+ T-cells expressing at least two markers among CD40L, IL-2, TNF-α, and IFN-γ upon stimulation with F, N and M2-1 peptide pools.

- Humoral response to the investigational RSV vaccine, pre-vaccination (Screening), post-Dose 1 (Day 30) and post-Dose 2 (Day 60 and Day 365):
  - Neutralizing antibody titers against RSV-A.
  - RSV F antibody concentrations.
- Humoral response to the investigational RSV vaccine, pre-vaccination (Screening), post-Dose 1 (Day 30) and post-Dose 2 (Day 60):
  - Palivizumab-competing antibody concentrations.

11.3. Tertiary endpoints

- CMI response profile (Th1, Th2, Th17) to the investigational RSV vaccine, pre-vaccination (Screening), post-Dose 1 (Day 30) and post-Dose 2 (Day 60 and Day 365).
  - CD3+/CD4+ and/or CD3+/CD8+ T-cells expressing at least one or any combination of immune marker(s) among CD40L, 41BB, IL-2, TNF-α, IFN-γ, IL-13 and IL-17 upon stimulation with F, N and M2-1 peptide pools.
- Any further exploratory immunology to detect disease-related or vaccine-related immune responses, such as but not limited to:
  - Anti-vector immunity: neutralization.

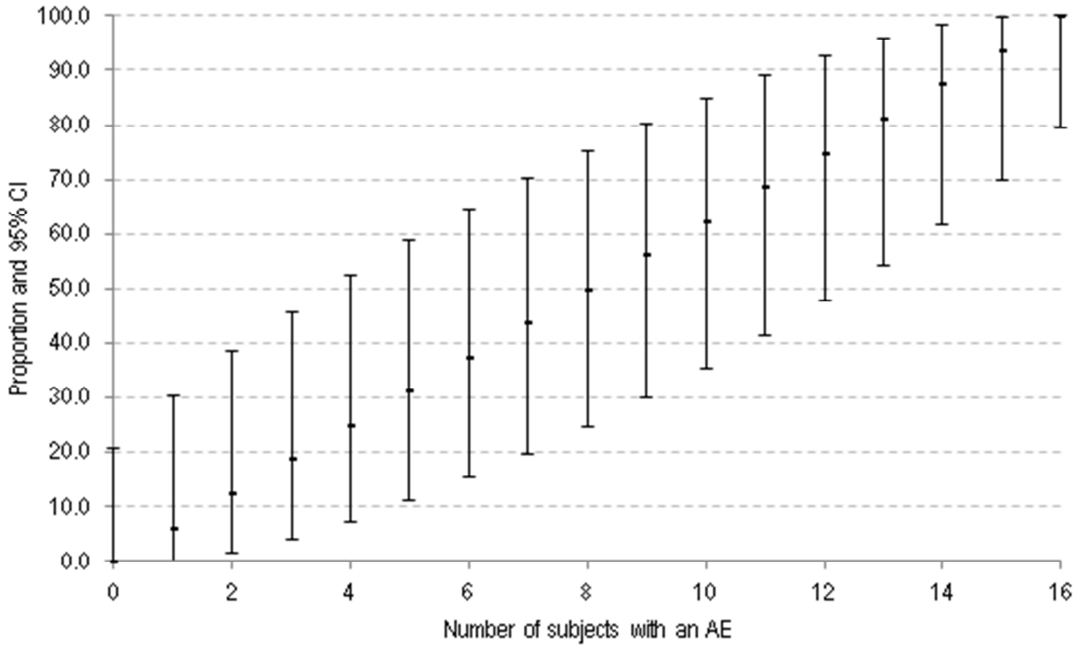
11.4. Determination of sample size

No hypothesis driven sample size calculation was conducted due to the nature of descriptive summary of the study objectives. The sample size of 32 subjects per dose cohort/Step, randomized 1:1 to vaccine or placebo, was based on conventional study designs for Phase 1/2 trials. Specifically, if no adverse event was observed among 16 RSV vaccinated subjects, the exact, two-sided 95% confidence interval (CI) would rule out an AE rate of 20% or more; if the incidence rate of AEs is more than 10%, there will still be more than an 80% chance of observing at least one subject with this AE. Combining all RSV vaccinees across all dose levels, there will be more than a 99% chance of observing at least one subject with this AE.

Furthermore, with 16 RSV vaccinees, the maximum width of a 95% CI for the proportion of subjects with adverse events among 16 vaccinees is about 50% (see Figure 5). **(Amended 10 December 2017)**

Figure 5 illustrates the precision on the percentage of subjects with symptoms following vaccination.

Figure 5 Exact 95% confidence interval on the percentage of subjects with adverse events following vaccination for 16 subjects per group



Taking the a priori safety concern of thrombocytopenia, both Frequentist and Bayesian approaches will be applied here to provide complementary assurance on the possibility to exclude a signal based on the accumulated safety data. The Frequentist approach examines the statistical power to rule out a significant decrease in platelet counts due to vaccination that would suggest a four-fold or greater increase in the risk of a Grade 3 or higher thrombocytopenia with 16 subjects per dose cohort. The Bayesian approach calculates the posterior predictive probability of not observing any subject with a Grade 3 or higher thrombocytopenia among the future 16 subjects by basing it on what we have observed in the first 16 subjects.

If we assume platelet counts in healthy infants between 12 and 23 months follow a normal distribution with a mean of 300,000 platelets/mm³ and a standard deviation of 75,000 platelets/mm³, then we would expect 0.04% of unvaccinated infants to have a count below 50000 platelets/mm³, which is the cut-off for a Grade 3 thrombocytopenia. To rule out a four-fold or greater increase, we would need to show that the mean platelet count does not fall below 190,000 platelets/mm³ with a standard deviation of 47,500. The sample size needed to rule out a 110,000 platelets/mm³ decrease due to vaccination with 84% power when vaccination has no effect on platelet count is 16 infants per dose cohort (8 per group). Sample size is based on a one-sided 0.025 test of non-inferiority using the Mann-Whitney statistic (PASS 12, Non-inferiority Tests for Two Means using Differences).

The Bayesian approach to predict the probability of no Grade 3 or higher thrombocytopenia in the future 16 subjects per dose cohort will be conducted by treating the mean of platelet counts μ as a random variable with a normal prior distribution $N(\mu_0, \sigma_0^2)$, where μ_0 and σ_0 will be determined by using laboratory normal

range. For this analysis the range of 150,000 to 450,000 platelets/mm³ will be the focus. Based on this laboratory range, μ_0 would be 300,000, a mean of the range, and σ_0 would be 75000, an approximate of a standard deviation of data sampled from this normal range. After 16 infants complete the two-dose vaccination schedule, a posterior distribution of μ will be calculated based on the prior of μ and the observed platelet counts from Day 31. The posterior predictive probability of no Grade 3 or higher thrombocytopenia in the future 16 subjects per dose cohort will be calculated, and sensitivity on the prior will be assessed.

After the first 16 infants of a dose cohort have completed the two-dose vaccination schedule, the 95% CI for the difference in mean platelets at Day 31 between vaccinees and placebo recipients will be calculated along with the posterior predictive probability of no Grade 3 or higher thrombocytopenia in the future 16 infants. If the lower boundary of the 95% CI is above 110,000 platelets/mm³ and the predictive probability is sufficiently high (e.g., 80% or more), the IDMC may recommend enrolling infants into the next dose cohort without completing enrolment in the current cohort. However, if a Grade 3 or higher thrombocytopenia occurs in a vaccinated infant at any time point or if there is a significant safety concern with another laboratory parameter such as hemoglobin or neutrophils, the IDMC may recommend continuing enrolment into the lower dose cohort. **(Amended 10 December 2017)**

11.5. Cohorts for Analyses

11.5.1. Total vaccinated cohort

The total vaccinated cohort (TVC) will include all subjects with at least one study vaccine administration documented.

A safety analysis based on the TVC will include all vaccinated subjects.

An immunogenicity analysis based on the TVC will include all vaccinated subjects for whom immunogenicity data are available.

The TVC analysis will be performed per treatment actually administered at Dose 1.

11.5.2. According-to-protocol cohort for analysis of immunogenicity

The ATP cohort for analysis of immunogenicity will be defined by timepoint and will consist of all subjects from the TVC who complied with eligibility criteria, study procedures up to the end of the study and had immunogenicity results in the epoch as described below.

More specifically, the ATP cohort for analysis of immunogenicity up to Visit 9 (Day 60)/at Visit 10 (Day 365) will include all evaluable subjects:

- Who met all eligibility criteria (i.e. no protocol violation linked to the inclusion/exclusion criteria, including age).

- Who received at least one dose of study vaccine.
- For whom the administration route and site of the vaccine was as according to protocol.
- Who received the vaccine according to protocol procedures.
- Who complied with the vaccination schedule, as specified in Table 5.
- Who did not receive a concomitant medication/product leading to exclusion from an ATP analysis, as described in Section 7.6.2, up to Visit 9 (Day 60)/up to Visit 10 (Day 365).
- Who complied with the timings of the post vaccination blood sampling for immune response evaluation, up to Visit 9 (Day 60) /at Visit 10 (Day 365), as specified in Table 5.
- For whom post-vaccination immunogenicity results are available for at least one assay up to Visit 9 (Day 60) /at Visit 10 (Day 365).

11.6. Derived and transformed data

11.6.1. Demography

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

11.6.2. Safety

For a given subject and the analysis of solicited AEs during the 7-day follow-up period after vaccination, missing or non-evaluable measurements will not be replaced. Therefore the analysis of the solicited symptoms based on the TVC will include only vaccinated subjects with documented safety data (i.e. symptom screen completed).

For analysis of unsolicited AEs, SAEs and for the analysis of concomitant medications, all vaccinated subjects will be considered. Subjects who did not report an event or concomitant medication will be considered as subjects without the event or the concomitant medication respectively.

11.6.3. Immunogenicity

Any missing or non-evaluable immunogenicity measurement will not be replaced:

- For the within-group assessment, the descriptive analysis performed for each assay at each timepoint will exclude subjects with a missing or non-evaluable measurement. Kinetics will be plotted on subjects with results available at all timepoints.

The geometric mean titers/concentrations (GMTs/GMCs) will be computed by taking the anti-logarithm of the arithmetic mean of the log10 transformed titers/concentrations.

A seronegative subject will be defined as a subject whose antibody titers/concentrations is below the cut-off value of the assay. A seropositive subject is a subject whose antibody titers/concentrations is greater than or equal to the cut-off value of the assay.

Vaccine response in terms of RSV neutralizing antibodies will be defined in the statistical analysis plan.

The description of the handling of data below the lower limit of quantification for GMC calculation and fold increase will be described in the statistical analysis plan.

11.6.4. RTI and LRTI

For the analysis of RTI and LRTI, all cases will be definitively classified as either RTI, LRTI or severe LRTI according to standardized case definitions (see Table 4) based on the available WHO case definitions, and the association to RSV infection will be assessed by quantitative PCR as primary analysis.

All confirmed LRTI will also be investigated for a panel of respiratory viruses (multiplex PCR; refer to Table 10), as a supplementary analysis of the occurrence of RSV-LRTI diagnosed upon the multiplex PCR.

For the analysis of RTI episode, a new RTI episode will be defined as any occurrence of cough, runny nose, blocked nose or difficulty breathing with an interval of at least 7 symptom free days since the last episode of RTI that was diagnosed (refer to Section 4.2 for the definition of start and end dates of RTI episode).

11.7. Analysis of demographics

The analysis of demographics will be performed on the TVC and on the ATP cohort for immunogenicity.

Demographic characteristics (age at vaccination in months, gender, race), weight, height or length, and vital signs, and cohort description will be summarized by group using descriptive statistics:

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

The distribution of subjects will be tabulated as a whole and per group.

Withdrawal status will be summarized by group using descriptive statistics:

- The number of subjects enrolled into the study as well as the number of subjects excluded from ATP analyses will be tabulated.
- The numbers of withdrawn subjects will be tabulated according to the reason for withdrawal.

11.8. Analysis of safety

11.8.1. Within groups assessment

The analysis will be performed on the TVC.

The percentage of subjects with at least one **local AE** (solicited and unsolicited), with at least one **general AE** (solicited and unsolicited) and with any AE during the 7-day or 30-day follow-up period will be tabulated with exact 95% CI after each vaccine dose and overall **(Amended 10 December 2017)**. The percentage of doses followed by at least one local AE (solicited and unsolicited), by at least one general AE (solicited and unsolicited) and by any AE during the 7-day or 30-day follow-up period will be tabulated, overall vaccination course, with exact 95% CI. The same computations will be done for Grade 3 AEs, for any AEs considered related to vaccination and for any Grade 3 AEs considered related to vaccination.

The percentage of subjects reporting each individual **solicited local AE** (any grade, Grade 2, Grade 3, resulting in a medically attended visit) and **solicited general AE** (any grade, Grade 2, Grade 3, any related, Grade 2 related, Grade 3 related, resulting in medically attended visit) during the 7-day follow-up period (Day 0-6) will be tabulated for each group after each vaccine dose and overall. Similarly, the percentage of doses followed by each individual solicited local and general AE and their sub-categories, will be tabulated, overall vaccination course, with exact 95% CI.

For fever, the number and percentage of subjects reporting fever by half degree (°C) cumulative increments during the 7-day follow-up period (Day 0-6) will be tabulated for each group after each vaccine dose and overall. Similar tabulations will be performed for any fever with a causal relationship to vaccination, Grade 3 (> 39.5°C) causally related fever and for any fever resulting in a medically attended visit. In addition, the prevalence of any and Grade 3 fever will be presented graphically over time after vaccination.

For each group and for each **hematology and biochemistry** parameter:

- The percentage of subjects having hematology and biochemistry results below or above the local laboratory normal ranges will be tabulated by timepoint.

  The maximum grading from Screening up to Visit 9 (Day 60) will be tabulated (Refer to Appendix C).

The percentage of subjects with **unsolicited** AEs within 30 days (Day 0-29) after each vaccine dose (overall doses) with its exact 95% CI will be tabulated by group and by 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 physician and the signs and AEs will be coded according to the MedDRA Dictionary for Adverse Reaction Terminology. Every verbatim term will be matched with the appropriate Preferred Term.

SAEs reported throughout the study and AE of specific interest will be described in detail.

The percentage of subjects using **concomitant medication** (any medication, any antipyretic and any antipyretic taken prophylactically, respectively) during the 7-day follow-up period (Day 0-6) and during the 30-day follow-up period (Day 0-29) will be summarized by group after each vaccine dose and overall.

11.8.2. Between groups assessment

Exploratory comparisons between the investigational RSV groups and the Placebo groups will be done in terms of the percentage of subjects, overall doses, reporting any Grade 2/3 AE during the 7-day follow-up period (Day 0-6) after vaccination, and/ or any fever >38.5°C during the 7-day follow-up period (Day 0-6) after vaccination, and/ or any vaccine-related SAE during the 7-day follow-up period (Day 0-6) after vaccination.

The standardized asymptotic 95% CI for the difference between the RSV groups and (minus) the Placebo groups will be computed.

11.9. Analysis of immunogenicity

The primary analysis will be performed on the ATP cohort for immunogenicity and, if in any group the percentage of vaccinated subjects with serological results excluded from the ATP cohort for analysis of immunogenicity is more than 10%, a second analysis will be performed on the TVC.

11.9.1. Within groups assessment

11.9.1.1. Analysis of secondary objectives

The following parameters will be summarized by group using descriptive statistics, at each timepoint during which blood samples are collected for CMI in investigational sites with a laboratory in proximity capable to perform WBS necessary for the CMI assay:

- Frequency of CD3+/CD4+ T-cells expressing at least two markers among CD40L, IL-2, TNF-α, and IFN-γ upon stimulation with F, N and M2-1 peptide pools.

For each group, at each timepoint that blood samples are collected for humoral immune response against the investigational RSV vaccine (neutralizing antibody titers against RSV-A, RSV F antibody concentrations, and palivizumab-competing antibody concentrations):

- GMTs/GMCs will be tabulated with 95% CI and represented graphically.
- Percentage of subjects above the seropositivity threshold will be tabulated with exact 95% CI.
- Antibody titers/concentrations will be displayed using reverse cumulative curves.

- The distributions of neutralizing antibody titers/concentrations will be tabulated.
- Percentage of responders in terms of neutralizing antibody titers will be tabulated with exact 95% CI.
- Individual post-vaccination versus pre-vaccination results will be plotted using scatter plots. Results of the Placebo groups will be used as a reference.
- Geometric mean of ratios of antibody titers/concentrations at each post-vaccination timepoint over pre-vaccination will be tabulated with 95% CI.
- Distribution of the fold increase of the neutralizing antibody titers will be tabulated by pre-vaccination titer category.
- The kinetics of individual antibody titer/antibody concentration results will be plotted as a function of time for subjects with results available at all timepoints.

11.9.1.2. Analysis of tertiary objectives

The cellular immune response will be further characterized using descriptive analysis of the frequency of CD3+/CD8+ T-cells and CD3+/CD4+ T-helper cell profile (Th1, Th2, Th17) expressing at least one or any combination of marker(s) among IL-2, CD40L, 41BB, TNF-α, IFN-γ, IL-13, and IL-17.

If available, any further exploratory immunology results (including, but not limited to anti-vector immunity) will be reported by group and timepoint using descriptive summary statistics.

11.10. Analysis of RTI and LRTI

The analysis will be performed on the TVC on two pooled groups (i.e. pooled RSV and pooled Placebo) separately for first and second RSV seasons and overall.

The proportion of subjects with at least one RSV-associated RTI (with 95 % CI) will be calculated. The same descriptive analysis will be performed for subjects with at least one RSV-associated LRTI and those with at least one RSV-associated severe LRTI.

As primary analysis, the assessment of RSV infection will be performed using the quantitative RT-PCR according to standardized case definitions (see Table 4) based on the available WHO case definitions.

Descriptive analyses (mean, median, min, max) of viral load assessed by the quantitative RT-PCR (RSV-A/B) of RSV-RTI, RSV-associated LRTI and severe LRTI cases will be tabulated. This analysis will also be done by study group.

The incidence rate of all-cause RTI (with 95% CI) will be calculated by group. The same descriptive analysis will be performed for all cause LRTI and all cause severe LRTI. These will also be presented for each viral etiology identified by multiplex PCR.

The incidence rate of asymptomatic RSV infections (with 95% CI) detected by the quantitative PCR (RSV-A/B) will be tabulated by group. Descriptive analyses (mean,

median, min, max) of viral load assessed by the quantitative RT-PCR (RSV-A/B) of those asymptomatic RSV infections will also be done by group.

11.11. Interpretation of analyses

Comparative analyses will be exploratory and should be interpreted with caution considering that there is no adjustment for multiplicity and that group sizes are small for these comparisons.

11.12. Conduct of analyses

Any deviation(s) or change(s) from the original statistical plan outlined in this protocol will be described and justified in the final study report.

11.12.1. Sequence of analyses

In preparation of the planned IDMC and iSRC evaluations, analyses of all available safety data (data that are as clean as possible) will be performed (see Section 9.10 for more information). These analyses will be done by an unblinded statistician outside GSK to maintain the study blind, and will be documented in a statistical analysis report. Only the outcome of the IDMC and iSRC reviews will be communicated to the RSV study team (no safety signal or safety signal). No clinical study report will be written.

The statistical analyses will be performed in 3 steps:

- A first analysis will be performed when all data up to Day 60 (i.e. data that are as clean as possible) are available. Additional safety data available at the time of this analysis will be described. At this point, the statistician will be unblinded (i.e. individual subject treatment assignments will be available) and the study will be conducted in a single-blind manner, with patients remaining blinded up to study end (Day 730). Summary results may unblind some specific subjects but no individual listings will be provided and the investigators will not have access to the treatment allocation up to study end (Day 730), except in case of emergency unblinding (see Section 9.8).
- An analysis will be performed when all data up to Visit 10 (Day 365; i.e. data that are as clean as possible) are available. No individual listings will be provided.
- The final analysis will be performed when all data up to study conclusion (Day 730) are available. An integrated clinical study report containing all data will be written and made available to the investigators at that time.

  If the data for tertiary endpoints become available at a later stage, (an) additional analysis/ analyses will be performed. These data will be documented in annex(es) to the study report and will be made available to the investigators at that time.

11.12.2. Statistical considerations for interim analyses

No interim analyses are planned.

12. ADMINISTRATIVE MATTERS

To comply with ICH GCP administrative obligations relating to data collection, monitoring, archiving data, audits, confidentiality and publications must be fulfilled.

12.1. electronic Case Report Form instructions

A validated GSK defined electronic data collection tool will be used as the method for data collection.

In all cases, subject initials will not be collected nor transmitted to GSK. Subject data necessary for analysis and reporting will be entered/transmitted into a validated database or data system. Clinical data management will be performed in accordance with applicable GSK standards and data cleaning procedures.

While completed eCRFs are reviewed by a GSK Biologicals' Site Monitor at the study site, omissions or inconsistencies detected by subsequent eCRF review may necessitate clarification or correction of omissions or inconsistencies with documentation and approval by the investigator or appropriately qualified designee. In all cases, the investigator remains accountable for the study data.

The investigator will be provided with a CD-ROM of the final version of the data generated at the investigational site once the database is archived and the study report is complete and approved by all parties.

12.2. Study Monitoring by GSK Biologicals

GSK will monitor the study to verify that, amongst others, the:

- Data are authentic, accurate, and complete.
- Safety and rights of subjects are being protected.
- Study is conducted in accordance with the currently approved protocol, any other study agreements, GCP and all applicable regulatory requirements.

The investigator and the head of the medical institution (where applicable) agrees to allow the monitor direct access to all relevant documents.

The investigator must ensure provision of reasonable time, space and qualified personnel for monitoring visits.

Direct access to all study-site related and source data is mandatory for the purpose of monitoring review. The monitor will perform an eCRF review and a Source Document Verification. By Source Document Verification we understand verifying eCRF entries by comparing them with the source data that will be made available by the investigator for this purpose.

The Source Documentation Agreement Form describes the source data for the different data in the eCRF. This document should be completed and signed by the site monitor and investigator and should be filed in the monitor's and investigator's study file. Any data item for which the eCRF will serve as the source must be identified, agreed and documented in the source documentation agreement form.

For eCRF, the monitor freezes completed and approved screens at each visit.

Upon completion or premature discontinuation of the study, the monitor will conduct site closure activities with the investigator or site staff, as appropriate, in accordance with applicable regulations, GCP, and GSK procedures.

12.3. Record retention

Following closure of the study, the investigator must maintain all site study records (except for those required by local regulations to be maintained elsewhere) in a safe and secure location. The records must be easily accessible, when needed (e.g. audit or inspection), and must be available for review in conjunction with assessment of the facility, supporting systems, and staff. Where permitted by applicable laws/regulations or institutional policy, some or all of these records can be maintained in a validated format other than hard copy (e.g. microfiche, scanned, electronic); however, caution needs to be exercised before such action is taken. The investigator must ensure that all reproductions are legible and are a true and accurate copy of the original and meet accessibility and retrieval standards, including re-generating a hard copy, if required. Furthermore, the investigator must ensure that an acceptable back-up of the reproductions exists and that there is an acceptable quality control procedure in place for making these reproductions.

GSK will inform the investigator/institution of the time period for retaining these records to comply with all applicable regulatory requirements. However, the investigator/institution should seek the written approval of the sponsor before proceeding with the disposal of these records. The minimum retention time will meet the strictest standard applicable to a particular site, as dictated by ICH GCP, any institutional requirements, applicable laws or regulations, or GSK standards/procedures.

The investigator/institution must notify GSK of any changes in the archival arrangements, including, but not limited to archival at an off-site facility, transfer of ownership of the records in the event the investigator leaves the site.

12.4. Quality assurance

To ensure compliance with GCP and all applicable regulatory requirements, GSK may conduct a quality assurance audit. Regulatory agencies may also conduct a regulatory inspection of this study. Such audits/inspections can occur at any time during or after completion of the study. If an audit or inspection occurs, the investigator and institution agree to allow the auditor/inspector direct access to all relevant documents and to allocate his/her time and the time of his/her staff to the auditor/inspector to discuss findings and any relevant issues.

12.5. Posting of information on publicly available clinical trial registers and publication policy

GSK assures that the key design elements of this protocol will be posted on the GSK website and in publicly accessible database(s) such as clinicaltrials.gov, in compliance with the current regulations.

GSK also assures that results of this study will be posted on the GSK website and in publicly accessible regulatory registry(ies) within the required time-frame, in compliance with the current regulations. The minimal requirement is to have primary endpoint summary results disclosed at latest 12 months post primary completion date and to have secondary endpoint disclosed at latest 12 months after the last subject last visit as described in the protocol.

As per EU regulation, summaries of the results of GSK interventional studies (phase I-IV) in pediatric population conducted in at least one EU member state will be posted on publicly available EMA registers within 6 months of end of study (as defined in the protocol) in the concerned EU member state. However, where, for scientific reasons detailed in the protocol, it is not possible to submit a summary of the results within 6 months in the concerned EU member state, the summary of results shall be submitted as soon as it is available. In this case, the protocol shall specify when the results are going to be submitted, together with a justification.

GSK also aims to publish the results of these studies in searchable, peer reviewed scientific literature and follows the guidance from the International Committee of Medical Journal Editors.

12.6. Provision of study results to investigators

Where required by applicable regulatory requirements, an investigator signatory will be identified for the approval of the study report. The investigator will be provided reasonable access to statistical tables, figures, and relevant reports and will have the opportunity to review the complete study results at a GSK site or other mutually-agreeable location.

GSK Biologicals will also provide the investigator with the full summary of the study results. The investigator is encouraged to share the summary results with the study subjects, as appropriate.

13. COUNTRY SPECIFIC REQUIREMENTS

In each participating country all children will be offered locally recommended vaccines for their age. These vaccines could be administered from Day 60 (Visit 9) onwards until the end of the study. Such vaccines will be provided by the investigator and where required will be sourced by GSK. The Investigator is responsible for reporting adverse events associated to the use of these vaccines according to local laws and regulations in addition to the safety reporting procedures foreseen in this study.

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APPENDIX A LABORATORY ASSAYS

Assay descriptions could be subjects to change, due to assay re-development and/or qualification.

Serostatus to RSV F and/or G protein

The selection of seropositive infants at screening will be based on ELISA with F and G protein coating antigens using a commercial kit (IBL kit).

Intracellular staining (ICS)

ICS has been used to assess CMI responses as previously described [Díez-Domingo, 2010; Moris, 2011].

Within 4 hours post phlebotomy, approximately 350 µL of undiluted whole blood are incubated at 37°C with the relevant antigen (in this case pools of 15-mer peptides overlapping by 11 amino acids and spanning the sequences of F, N, and M2-1 proteins), negative control (medium), or positive control (staphylococcal enterotoxin B) for 2 hours in the presence of anti-CD28 and anti-CD49d antibodies. Brefeldin A, a protein secretion inhibitor, is then added and the mix is incubated overnight. Red cells are then lysed and white cells washed, fixed, cryopreserved, and stored until analysis by cytometry.

Once ready for cytometry analysis, white cells are thawed in batches, washed, and incubated with fluorochrome-conjugated antibodies specific to CD4 and CD8 surface markers. Then, cells are permeabilized and stained with fluorochrome-conjugated antibodies specific for the following immune markers: IL-2, IFN-γ, TNF-α, CD40L, IL-13, IL-17, and 41BB. After washing, acquisition of data is performed via a flow cytometer.

The results are expressed as the frequency of CD4+ or CD8+ T-cells expressing, per million of CD4+ or CD8+ T-cells:

- CD3+/CD4+ T-cells expressing at least two markers among CD40L, IL-2, TNF-α, and IFN-γ (poly-functional CD4+ T-cells).
- CD3+/CD4+ and/or CD3+/CD8+ T-cells expressing at least one or any combination of immune marker(s) among CD40L, 41BB, IL-2, TNF-α, IFN-γ, IL-13 and IL-17 (for determination of Th profile).

Neutralization assay

The serum neutralization assay is a functional assay that measures the ability of serum antibodies to neutralize RSV entry and replication in a host cell line.

First, virus neutralization is performed by incubating a fixed amount of RSV-A long strain (ATCC No. VR-26) with serial dilutions of the test serum. Then, the serum-virus mixture is transferred onto a monolayer of Vero cells (African Green Monkey, kidney, Cercopitheus aethiops, ATCC CCL-81) and incubated for three days to allow infection of Vero cells by non-neutralized viruses and the formation of plaques in the cell monolayer.

Following the fixation period, RSV-infected cells are detected using a primary antibody directed against RSV (anti-RSV IgG) and a secondary antibody conjugated with fluorescein isothiocyanate, allowing the visualization of plaques by immunofluorescence. Viral plaques are counted using an automated microscope coupled to an image analyzer (Scanlab system with Axiovision software). For each serum dilution, a ratio, expressed as a percentage, is calculated between the number of plaques at that dilution and the number of plaques in the virus control wells (no serum added). The serum neutralizing antibody titer is expressed in ED60 (Estimated Dilution 60) and corresponds to the inverse of the interpolated serum dilution that yields a 60% reduction in the number of plaques compared to the virus control wells as described by others [Barbas, 1992; Bates, 2014].

ELISA

- Anti-RSV protein F ELISA

  The anti-F protein IgG ELISA is an indirect ELISA allowing the detection and the quantitation of specific IgG antibodies directed against the RSV F protein in human serum samples. **(Amended 10 December 2017)**

- Palivizumab competitive assay

  Palivizumab monoclonal antibody (Synagis) is used as a passive treatment that protects against RSV infection by binding to the antigenic site II epitope of the RSV F antigen.

  First, F protein antigens purified from CHO expression system are coated onto 96-well microplates. Then, after a washing and a blocking step, serial two-fold dilutions of test sera, positive control serum, and palivizumab antibody reference standard are added in sequence with competitor antibodies (horseradish peroxidase-conjugated palivizumab) and incubated to allow specific binding of antibodies directed against the F protein antigens. If palivizumab-like antibodies are present in serum samples, they will compete with the horseradish peroxidase-conjugated palivizumab antibodies for binding to the F protein coated antigen. After a washing step, the horseradish peroxidase substrate solution (TMB/H_2O_2) is added and a colored product develops in a manner that is inversely proportional to the amount of palivizumab-like antibodies contained in the test serum. The color is quantified by reading the optical densities at 450-620 nm using a spectrophotometer. Antibody concentrations of individual serum and control samples are determined after interpolation from the ELISA standard curve using a four-parameter equation and are expressed as palivizumab-equivalent antibodies in microgram per millilitre (µg/mL).

PCR

- Quantitative PCR able to discriminate RSV-A and RSV-B subtypes:

  Briefly, RSV A and RSV B RNAs extracted from the nasal swabs are detected in a duplex PCR format using specific amplification primers and fluorescent probes designed in the RSV N gene, encoding the RSV nucleocapsid protein. The process involves nucleic acids extraction, conversion of RNA to complementary deoxyribonucleic acid by reverse transcription and detection by real- time PCR reaction using a calibration curve (absolute quantitation). The RSV viral load is reported as copies of RSV RNA per mL of sample.

- Qualitative multiplex PCR for detection of a panel of viruses.

  A qualitative PCR multiplex assay is used for the detection and identification of multiple respiratory virus nucleic acids in nasal swabs from individuals suspected of respiratory tract infections. The following virus types and subtypes can be identified in the assay:

  – Influenza A virus (Flu A)
  – Influenza B virus (Flu B)
  – Human Influenza A virus subtype H1 (Flu A-H1)
  – Human Influenza A virus subtype H3 (Flu A-H3)
  – Human Influenza A virus subtype H1pdm09 (Flu A-H1pdm09)
  – Human respiratory syncytial virus A (RSV A)
  – Human respiratory syncytial virus B (RSV B)
  – Human adenovirus (AdV)
  – Human metapneumovirus (MPV)
  – Human enterovirus (HEV)
  – Human parainfluenza virus 1 (PIV1)
  – Human parainfluenza virus 2 (PIV2)
  – Human parainfluenza virus 3 (PIV3)
  – Human parainfluenza virus 4 (PIV4)
  – Human bocavirus 1/2/3/4 (HBoV)
  – Human rhinovirus A/B/C (HRV)
  – Human coronavirus 229E (229E)
  – Human coronavirus NL63 (NL63)
  – Human coronavirus OC43 (OC43)

  Following total nucleic acids extraction, viruses are detected by multiplex real-time RT-PCR assays targeting the above mentioned viruses. A comparative analysis of the fluorescence intensities of each target is performed to detect the viruses present in the sample.

APPENDIX B CLINICAL LABORATORIES

Table 23 GSK Biologicals' laboratories

Laboratory	Address
GSK Biologicals Clinical Laboratory Sciences, Rixensart	Biospecimen Reception - B7/44 Rue de l'Institut, 89 - B-1330 Rixensart - Belgium
GSK Biologicals Clinical Laboratory Sciences, Wavre-Nord Noir Epine	Avenue Fleming, 20 - B-1300 Wavre - Belgium

Table 24 Outsourced laboratories

Laboratory	Address
NÉOMED-LABS Inc.	525, Cartier Ouest Laval, Quebec Canada H7V 3S8
Q^2 Solutions Clinical Trials (US)	27027 Tourney Road, Suite 2E Valencia, CA 91355 USA

APPENDIX C TOXICITY GRADING FOR HEMATOLOGY AND BIOCHEMISTRY PARAMETERS

Table 25 Toxicity grading scales for hematology and biochemistry parameters applicable for this study (Amended 10 December 2017)

Component	Grade 1	Grade 2	Grade 3	Grade 4
Hemoglobin (g/dL)	9.0 to < 10.5	8.0 to < 9.0	7.0 to < 8.0	< 7.0
Leukocytes decreased (cell/mm^3)	2500 to < 3500	1500 to < 2500	1000 to < 1500	< 1000
Absolute neutrophil count decreased (cell/mm^3)	***1000 to < 1300***	***750 to < 1000***	***500 to < 750***	***< 500***
Absolute lymphocyte count decreased (cell/mm^3)	***600 to < 650***	***500 to < 600***	***350 to < 500***	***< 350***
Platelets decreased (cell/mm^3)	75000 to < 150000	50000 to < 75000	25000 to < 50000	< 25000
Alanine Aminotransferase (increase by factor)	1.1 to < 2.0 xULN	2.0 to < 3.0 xULN	3.00 to ≤ 8.0 xULN	> 8.0 xULN
Aspartate Aminotransferase (increase by factor)	1.1 to < 2.0 xULN	2.0 to < 3.0 xULN	3.00 to ≤ 8.0 xULN	> 8.0 xULN
Creatinine (mg/dL)	0.6 to < 0.9	0.9 to < 1.2	1.2 to ≤ 1.5	> 1.5

Grading scale adapted from [Division of AIDS, 2003], [Division of AIDS, 2004] and [Division of AIDS, 2007].
ULN: upper limit of normal

APPENDIX D AMENDMENTS AND ADMINISTRATIVE CHANGES TO THE PROTOCOL

GlaxoSmithKline Biologicals SA Vaccines R &D Protocol Administrative Change 1	
eTrack study number and Abbreviated Title	204838 (RSV PED-002)
IND number	16999
EudraCT number	2016-000117-76
Administrative change number:	Administrative change 1
Administrative change date:	06 July 2016
Co-ordinating author:	PPD (Project Manager Scientific Writer, Keyrus Biopharma contractor for GSK Biologicals)

Rationale/background for changes:

- The IND number has been added on the protocol cover page, the Sponsor Signatory Approval page and the Investigator agreement page.
- Change in study personnel has been mentioned on the protocol cover page.

Amended text has been included in *bold italics* and deleted text in ~~strikethrough~~ in the following sections:

- On the protocol cover page, the Sponsor Signatory Approval page and the Investigator agreement page, IND number has been added:

Investigational New Drug (IND) number	***16999***

- On the protocol cover page, a contributing author has been added.

Contributing authors (continued)	PPD ***(Global Regulatory Representative)***

In the List of abbreviations, definition of the abbreviation "IND" has been added:

IND:	**Investigational New Drug**

GlaxoSmithKline Biologicals SA

Vaccines R &D
Protocol Administrative Change 2

eTrack study number and Abbreviated Title	204838 (RSV PED-002)
IND number	16999
EudraCT number	2016-000117-76
Administrative change number:	Administrative change 2
Administrative change date:	13 September 2016
Co-ordinating author:	PPD (Project Manager Scientific Writer, Keyrus Biopharma contractor for GSK Biologicals)

Rationale/background for changes:

- The GSK Biologicals' Contact information for Emergency Unblinding for US and Canada have changed. This protocol administrative change provides the new numbers.
- Change in study personnel has been mentioned on the protocol cover page.

Amended text has been included in *bold italics* and deleted text in ~~strikethrough~~ in the following sections:

- On the **protocol cover page**, the following contributing authors have been added.

Contributing authors (continued)

- PPD ***(Vaccine Supply Coordinator)***
- PPD ***(Oversight Data Manager, Keyrus Biopharma contractor for GSK Biologicals)***

- In **Section 9.8. Emergency unblinding**, the GSK Biologicals' Contact information for Emergency Unblinding for US and Canada have been changed.

GSK Biologicals' Contact information for Emergency Unblinding **24/24 hour and 7/7 day availability**
GSK Biologicals' Central Safety Physician: Outside US: PPD (GSK Biologicals Central Safety Physician on-call) For US: PPD PPD (GSK Biologicals Central Safety Physician on-call) **GSK Biologicals' Central Safety Physician Back-up:** Outside US: PPD US only: PPD PPD **Emergency Unblinding Documentation Form transmission:** Outside US: Fax: PPD or PPD US only: Fax: PPD

GlaxoSmithKline Biologicals SA

Vaccines R &D

Protocol Amendment 1

eTrack study number and Abbreviated Title:	204838 (RSV PED-002)
IND number:	16999
EudraCT number:	2016-000117-76
Amendment number:	Amendment 1
Amendment date:	16 January 2017
Co-ordinating author:	PPD (Project Manager Scientific Writer, Keyrus Biopharma contractor for GSK Biologicals)

Rationale/background for changes:

- The Spanish competent authorities (Agencia Española de Medicamentos y Productos Sanitarios [AEMPS]) requested the Sponsor to extend the long term medical follow-up for one additional year. The purpose of this amendment is to respond to this request by increasing the follow-up of the subjects from one year to two years. This will include the collection of adverse events (adverse events of specific interest [RSV-LRTI] and serious adverse events), the surveillance for respiratory tract infection, difficulty of breathing and wheezing.
- The Italian competent authorities (Agenzia Italiana del Farmaco [AIFA]) requested that, in addition to the 60 minutes observation planned per protocol between each infants receiving ChAd155-RSV vaccination, the Sponsor monitors potential hypersensitivity reactions for a longer period following both vaccine administrations (one month apart) in a sufficient number of infants before another one can be vaccinated with the same dose. The Sponsor agreed to put in place a 48-hour observation period after the first and second doses administered to the first eight infants enrolled in each step, before continuing vaccination of more infants with a minimum interval of 60 minutes. The purpose of this amendment is to respond to this request.
- Following the request from investigators, the screening period has been increased from 15 days (Day -14 to Day 0) to 30 days (Day -29 to Day 0) in order to improve recruitment of subjects.
- Following the request from investigators, the following exclusion criteria have been clarified:
  - History of wheezing.
  - History of chronic cough.
- The satisfactory outcome of the review by an Independent Data Monitoring Committee (IDMC) of safety data from study 201974 (RSV PED-001) in healthy adults has been added to the rationale for the choice of study population.

- The holding rule for hospitalization due to any local or general solicited AE has been restricted to any local or general solicited AE that cannot reasonably be attributed to a cause other than vaccination in order to exclude irrelevant cases.
- In addition, possibility to assess RSV infection using WHO case definition, has been added to this protocol.
- In addition, some typos have been corrected and other administrative changes have been made throughout the protocol in order to harmonize wording across the different RSV project documents and to add clarity.
- Finally, changes in study personnel have been included on the protocol cover page.

Amended text has been included in *bold italics* and deleted text in ~~strikethrough~~ in the following sections:

On **the protocol cover page**, the following changes have been made:

Contributing authors

- PPD ***(Lead Statistician)***
- PPD ***(Project Statistician)***
- PPD ***(Study Delivery Lead)***
- PPD ***(Vaccine Supply Coordinator, Aixial contractor for GSK Biologicals)***
- PPD (Oversight Data Manager, ~~Keyrus Biopharma contractor for GSK Biologicals~~)
- PPD ***(Global Regulatory Representative)***

In the **Synopsis**, the following changes have been made:

Rationale for the study and study design

- **Study population**: The immunogenicity, safety and reactogenicity of the pediatric candidate RSV vaccine (ChAd155-RSV vaccine) are currently being evaluated in healthy adult subjects aged 18 to 45 years (study 201974 [RSV PED-001]). The ChAd155-RSV vaccine is being developed for the active immunization of infants as from 6 weeks of age. To allow age de-escalation from the adult population to the targeted population and to not enroll and administer the ChAd155-RSV vaccine to older children who are beyond the age of severe RSV disease and would not benefit from this vaccine, the present study will be performed on RSV-seropositive infants aged 12 to 17 months, ~~pending satisfactory~~ ***after the*** safety profile of the ChAd155-RSV vaccine in adults (study 201974 [RSV PED-001; NCT02491463]) ***was evaluated as satisfactory by an Independent Data Monitoring Committee (IDMC)***. The present study will provide critical information on

the safety and immunogenicity profile of the ChAd155-RSV vaccine before a subsequent trial in seronegative infants.

- **Study blinding:** Given the different appearance and storage conditions of the investigational RSV vaccine and placebo, double blinding is not possible and the study will be conducted in an observer-blind manner.

  When all data up to Day 60 are available, a statistical analysis will be performed. This analysis may lead to the unblinding of some subjects. As a consequence, after Day 60, the study cannot be considered as observer-blind, but will be conducted in a single-blind manner, with subjects' parent(s)/ legally acceptable representative (LAR[s]) remaining blinded up to study end (~~Day 365~~***Day 730***). The investigators will not have access to the treatment allocation up to study end (~~Day 365~~***Day 730***).

- **Staggered design with 3 steps:**

  Vaccination will be limited to a maximum of 10 infants per day. In the same investigational center, infants will be vaccinated sequentially, separated by a minimum interval of 60 minutes to allow monitoring of any acute events (e.g. hypersensitivity reaction). ***In addition, to monitor delayed potential reactions following vaccination, there will be a 48-hour observation period following Dose 1 and Dose 2 administration to the first eight infants (corresponding to three to five ChAd155-RSV vaccine recipients) in each Step (1, 2, and 3) before continuing vaccination of more infants (i.e. from the ninth infant). From the ninth infant, infants will be vaccinated sequentially, separated by a minimum interval of 60 minutes. Refer to the study procedures manual (SPM) for more details about the 48-hour observation period after the first eight subjects vaccinated in each Step.***

  Within each step, an internal Safety Review Committee (iSRC) will review accumulated safety data three weeks after the start of vaccination and then about every three weeks (until the I~~ndependent Data Monitoring Committee [~~IDMC~~]~~ reviews the data).

Objectives	**Secondary** • To evaluate the safety of two IM doses of three dose levels of the RSV investigational vaccine when administered according to a 0, 1-month schedule from study start (Day 0) up to study conclusion (Day ~~365~~***730***) in RSV-seropositive infants aged 12 to 17 months. • To evaluate the occurrence of RSV respiratory tract infections in RSV-seropositive infants from Visit 1 (Day 0, after Dose 1) up to study conclusion (Day ~~365~~***730***).
Study design	• Duration of the study: approximately ~~12~~ ***24*** months: • Epoch 002: follow-up starting at Visit 10 (Day 365) and ending at Visit ~~10~~ ***11*** (Day ~~365~~ ***730***). • End of Study: Last testing results released of samples collected at Visit ~~10~~ ***11*** (Day ~~365~~ ***730***). • Sampling schedule: – Blood samples for **biochemistry** will be taken from all infants at Screening (~~14~~ ***29*** days before first vaccination to Day 0) and on Day 7, Day 30, Day 37, and Day 60. Blood samples for **hematology** will be taken from all infants at Screening (~~14~~ ***29*** days before first vaccination to Day 0) and on Day 1, Day 7, Day 30, Day 31, Day 37, and Day 60. A clinical history and examination with special attention given to detection of spontaneous bleeding or easy bruising and evidence of bruising or petechiae will be performed on Day 1, Day 3 and Day 7 after each vaccination (i.e. Day 1, Day 3, Day 7, Day 31, Day 33, and Day 37). On Day 1, Day 7, Day 31, and Day 37, special attention will also be given to the detection of petechiae or evidence of petechiae induced by the tourniquet. Further testing may be required to investigate a finding or guide subject management based on the investigators clinical judgment. Subjects' parent(s)/LAR(s) will be instructed to contact the investigator/study staff if their child presents symptoms of spontaneous bleeding or easy bruising following vaccination. • Surveillance for RSV-RTI, difficulty in breathing and wheezing episodes.

Surveillance period will be carried out from Visit 1 (after Dose 1) until Visit ~~10~~ ***11*** (Day ~~365~~***730***). In order to detect asymptomatic RSV-RTI, monthly nasal swabs for analysis at sponsor laboratory will be performed for all subjects during the RSV season. In order to timely detect RSV-RTI and to ensure cases are timely captured by the study sites, both active and passive surveillance will be conducted:

Case definition

During the analysis of the study, all cases identified during the surveillance of RSV-RTI will be definitively classified as either RTI, LRTI or severe LRTI according to the case definitions described in Synopsis Table 4. ***The analysis may also be done using the available World Health Organization (WHO) case definitions for RTI, LRTI and severe LRTI.***

Endpoints

Secondary

- Occurrence of SAEs from study start (Day 0) up to study conclusion (Day ~~365~~***730***) in all subjects.
- Occurrence of RSV-LRTI (AE of specific interest) as from Dose 1 administration up to study conclusion (Day ~~365~~***730***) in all subjects.
- Occurrence of RSV-RTI, RSV-LRTI, severe RSV-LRTI (according to case definitions) as from Dose 1 administration up to study conclusion (Day ~~365~~***730***) in all subjects.

In **Section 1.2.2.1 Study population**, the following changes have been made:

- The immunogenicity, safety and reactogenicity of the pediatric candidate RSV vaccine (ChAd155-RSV vaccine) are currently being evaluated in healthy adults aged 18 to 45 years (study 201974 [RSV PED-001; NCT02491463]). The ChAd155-RSV vaccine is being developed for the active immunization of infants as from 6 weeks of age. To allow age de-escalation from the adult population to the targeted population and to not enroll and administer the ChAd155-RSV vaccine to older children who are beyond the age of severe RSV disease and would not benefit from this vaccine, the present study will be performed on RSV-seropositive infants aged 12 to 17 months, ~~pending satisfactory~~ ***after the*** safety profile of the ChAd155-RSV vaccine in adults (study 201974 [RSV PED-001; NCT02491463]) ***was evaluated as satisfactory by an Independent Data Monitoring Committee (IDMC)***. The present study will provide critical information on the safety and immunogenicity profile of the ChAd155-RSV vaccine before a subsequent trial in seronegative infants.

In **Section 1.2.2.2 RSV serostatus using IBL International kit and choice of study population**, the following changes have been made:

- Despite the assay characterization performed by the manufacturer, a potential lack of sensitivity of the IBL International kit compared to a RSV neutralization assay has been reported [Malkin, 2013]. GSK considers that ***the specificity and sensitivity characteristics of the assay indicate a reduced*** ~~a lower sensitivity reduces the~~ risk of enrolling RSV-seronegative infants ***identified RSV-seropositive in the assay*** (i.e. false ~~RSV-sero~~positive ~~infants~~ ***results***~~) and that this is acceptable in this screening context~~.

In **Section 1.2.4.1 Staggered design with 3 steps**, the following change has been made:

- Vaccination will be limited to a maximum of 10 infants per day. In the same investigational center, infants will be vaccinated sequentially, separated by a minimum interval of 60 minutes to allow monitoring of any acute events (e.g. hypersensitivity reaction). ***In addition, to monitor delayed potential reactions following vaccination, there will be a 48-hour observation period following Dose 1 and Dose 2 administration to the first eight infants (corresponding to three to five ChAd155-RSV vaccine recipients) in each Step (1, 2, and 3) before continuing vaccination of more infant (i.e. from the ninth infant). From the ninth infant, infants will be vaccinated sequentially, separated by a minimum interval of 60 minutes. Refer to the study procedures manual (SPM) for more details about the 48-hour observation period after the first eight subjects vaccinated in each Step.***
- Within each step, an internal Safety Review Committee (iSRC) will review accumulated safety data three weeks after the start of vaccination and then about every three weeks (until the ~~Independent Data Monitoring Committee [~~IDMC~~]~~ reviews the data). Ad-hoc iSRC meetings may be held should safety concerns warrant a safety review. Refer to Section 9.10.2 for more information about iSRC.

In **Section 1.2.4.2 Rationale for monitoring of hematology parameters**, the following changes have been made:

- Emphasis on "minimal risk" is imperative for the protection of the pediatric population participating to research studies where blood sample volume limits is a critical parameter. To stay within blood sampling limits that are consistent with physiological "minimal risk" in this young and vulnerable population, but also to minimize any development of needle aversion, no blood draw will be performed for immunological assays and for biochemistry tests at Day 1 and none for hematology and biochemistry tests at Day 3. Blood samples collected within the screening window ~~(limited to maximum 14 days)~~ will serve as baseline for the immunological analysis and for the hematology and biochemistry check-up before vaccination. Consequently, the maximum blood draw volumes collected per protocol have been restricted to 9.0 mL within 24 hours and 32.4 mL over eight weeks (refer to Table 5 and Table 7). Of note, it is considered that blood sample volumes ranging from 1 to 5% of total blood volume within 24 hours and up to 10% of total blood volume over 8 weeks are consistent with the limited evidence available on the minimal physiological risk for healthy children [Howie; 2011]. The total blood volume of a child is around 75 to 80 mL/kg (85 to 105 mL/kg in the neonatal period). Considering a girl of 12 months of age with a low weight of 7.3 kg (5^{th} percentile [WHO; 2006]), the minimal individual total blood volume in the study population would be 547.5 mL and therefore the safer limits of blood draw volumes would range from 5.5 to 27.4 mL within 24 hours and be limited to 54.7 mL over 8 weeks. To minimize any development of needle aversion, pain relief by means of a topical local anesthetic may be offered to infants prior to any blood sampling requested by the protocol at the discretion of the investigator.

In **Section 1.2.5 Rationale for study blinding**, the following changes have been made:

- When all data up to Day 60 are available, a statistical analysis will be performed. This analysis may lead to the unblinding of some subjects. As a consequence, after Day 60, the study cannot be considered as observer-blind, but will be conducted in a single-blind manner, with subjects' parent(s)/ LAR(s) remaining blinded up to study end (Day ~~365~~***730***). The investigators will not have access to the treatment allocation up to study end (Day ~~365~~***730***).

In **Section 2.2 Secondary objectives**, the following changes have been made:

- To evaluate the safety of two IM doses of three dose levels of the RSV investigational vaccine when administered according to a 0, 1-month schedule from study start (Day 0) up to study conclusion (Day ~~365~~***730***) in RSV-seropositive infants aged 12 to 17 months.
- To evaluate the occurrence of RSV respiratory tract infections in RSV-seropositive infants from Visit 1 (Day 0, after Dose 1) up to study conclusion (Day ~~365~~***730***).

In **Section 3 Study Design Overview**, the following changes have been made:

Figure 1 Study design

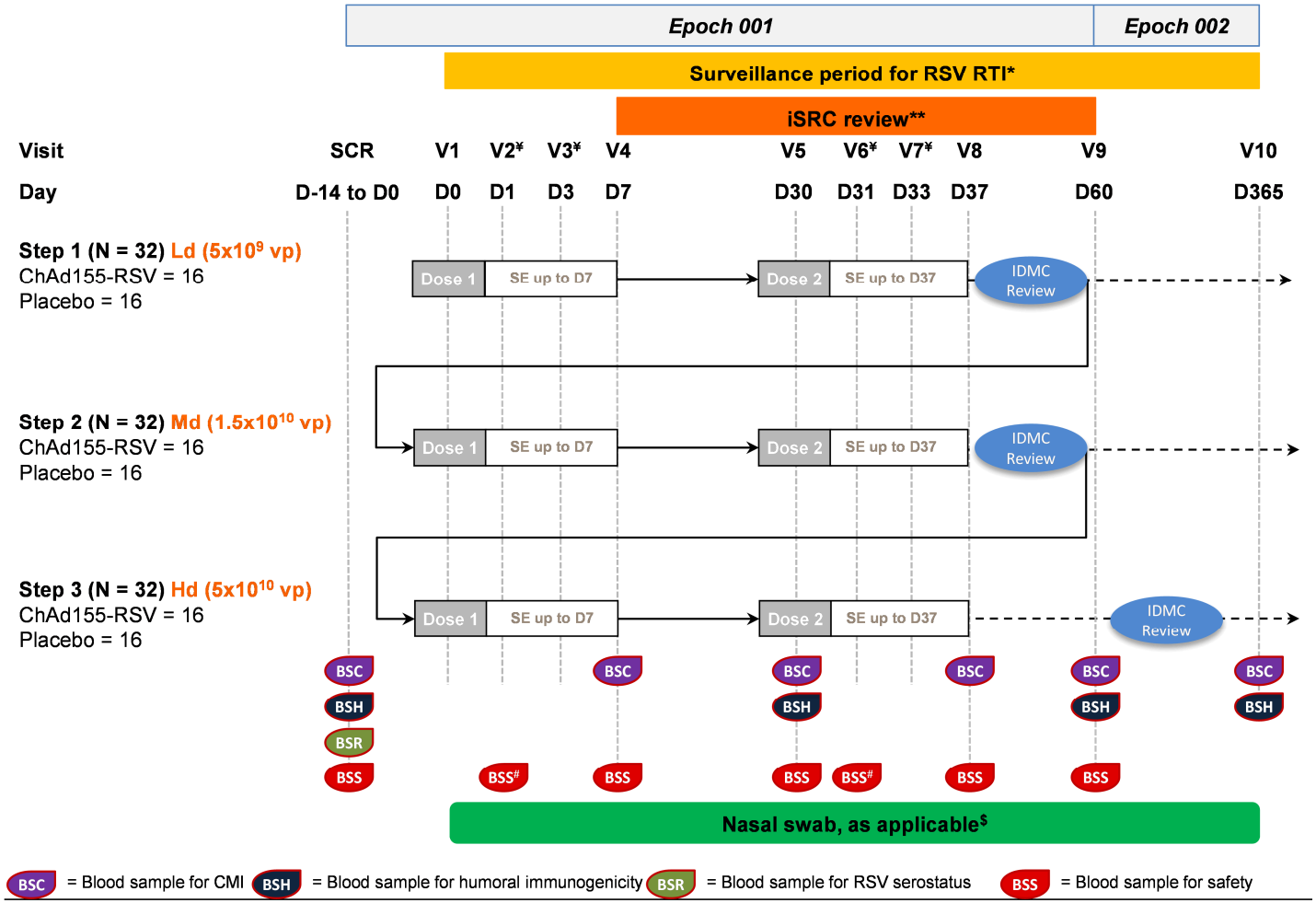
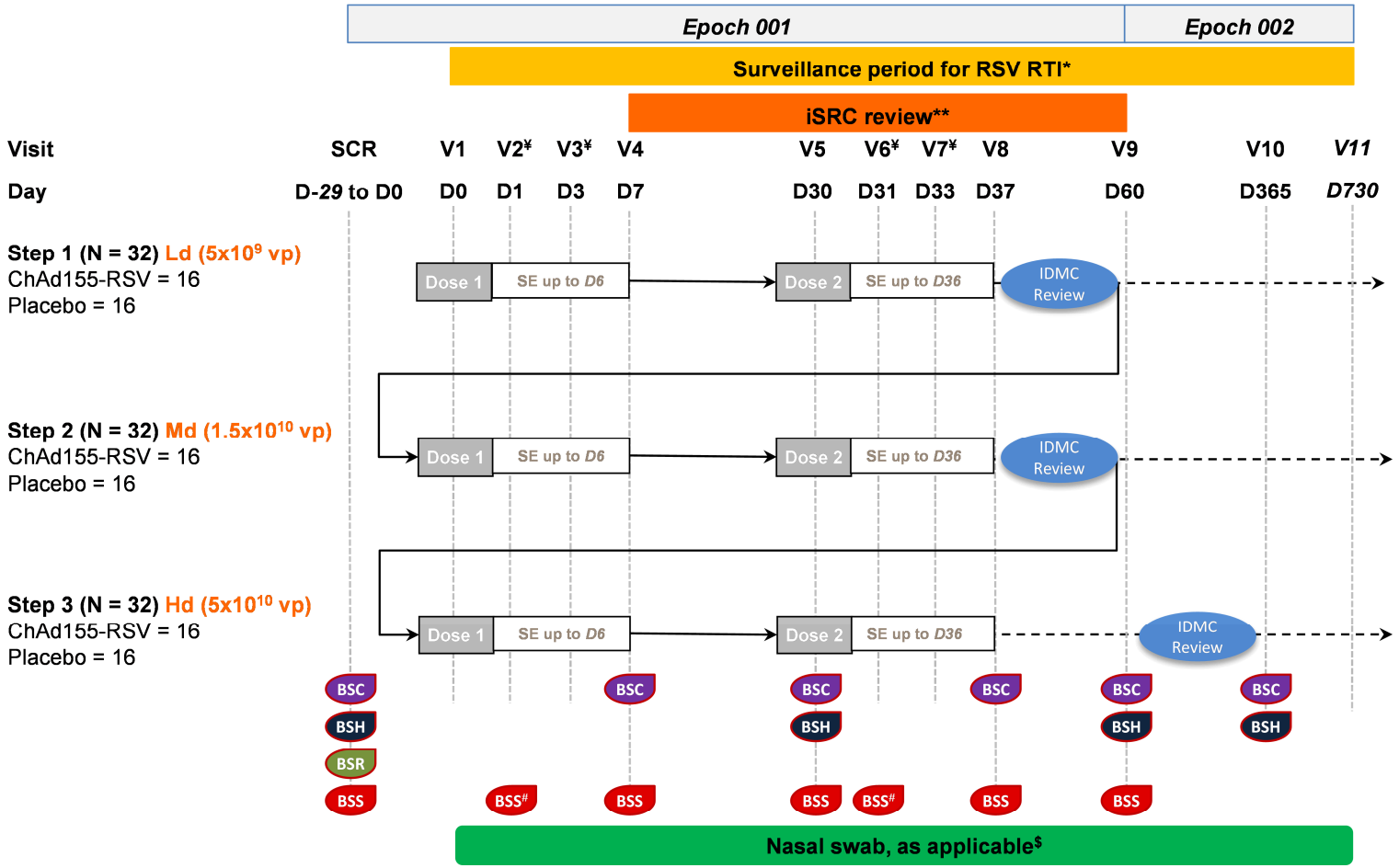


Figure 1 Study design



¥ Visit 2 (Day 1), Visit 3 (Day 3), Visit 6 (Day 31), ~~and~~ Visit 7 (Day 33), ***and Visit 11 (Day 730)*** may take place in the subject's home or at the investigators clinical facility as appropriate

- Duration of the study: approximately ~~12~~ ***24*** months:
  - Epoch 002: follow-up starting at Visit 10 (Day 365) and ending at Visit ~~10~~ ***11*** (Day ~~365~~ ***730***).
- End of Study: Last testing results released of samples collected at Visit ~~10~~ ***11*** (Day ~~365~~ ***730***).
- Sampling schedule:
  - Blood samples for **biochemistry** will be taken from all infants at Screening (up to ~~14~~ ***29*** days before first vaccination to Day 0) and on Day 7, Day 30, Day 37, and Day 60 (refer to Table 11 for the list of parameters to be tested). Blood samples for **hematology** will be taken from all infants at Screening (up to ~~14~~ ***29*** days before first vaccination to Day 0) and on Day 1, Day 7, Day 30, Day 31, Day 37, and Day 60 (refer to Table 11 for the list of parameters to be tested). A clinical history and examination with special attention given to detection of spontaneous bleeding or easy bruising and evidence of bruising or petechiae will be performed on Day 1, Day 3 and Day 7 after each vaccination (i.e. Day 1, Day 3, Day 7, Day 31, Day 33, and Day 37). On Day 1, Day 7, Day 31, and Day 37, special attention will also be given to the detection of petechiae or evidence of petechiae induced by the tourniquet. Further testing may be required to investigate a finding or guide subject management based on the investigators clinical judgment. Subjects' parent(s)/LAR(s) will be instructed to contact the investigator/study staff if their child presents symptoms of spontaneous bleeding or easy bruising following vaccination. Refer to Section 6.6.11.2 and Figure 2 for information about the re-testing of samples in case of any Grade 1 abnormality with potential clinical relevance or any ≥ Grade 2 abnormality.
- Surveillance for RSV-RTI, difficulty in breathing and wheezing episodes.

  Surveillance period will be carried out from Visit 1 (after Dose 1) until Visit ~~10~~ ***11*** (Day ~~365~~***730***). In order to detect asymptomatic RSV-RTI, monthly nasal swabs for analysis at sponsor laboratory will be performed for all subjects during the RSV season. In order to timely detect RSV-RTI and to ensure cases are timely captured by the study sites, both active and passive surveillance will be conducted:

In **Section 4.1 RTI case definitions**, the following change has been made:

- During the analysis of the study, all cases identified during the surveillance of RSV-RTI will be definitively classified as either RTI, LRTI or severe LRTI according to the case definitions described in Table 4. ***The analysis may also be done using the available World Health Organization (WHO) case definitions for RTI, LRTI and severe LRTI.***

In **Section 4.2.1 Start date of the RTI episode**, the following change has been made:

- The start date of the RTI episode is defined as the point at which the first symptoms of cough, runny nose, blocked nose, ***wheezing,*** or difficulty breathing were observed.

In **Section 4.2.2 End date of the RTI episode**, the following change has been made:

- The end date of the RTI episode is defined as the point at which the child is considered symptom-free of cough, runny nose, blocked nose, ***wheezing,*** or difficulty breathing.

In **Section 5.3 Exclusion criteria for enrolment**, the following changes have been made:

- History of ***recurrent*** wheezing. ***(Wheezing should have been verified on auscultation by doctor).***
- History of chronic cough ***(4 weeks or more duration)***.

In **Section 6.2.2.2.1 Study group and treatment number allocation**, the following change has been made:

- When SBIR is not available, please refer to the SBIR user guide or the ~~Study Procedures Manual (~~SPM~~)~~ for specific instructions.

In **Section 6.3 Method of blinding**, the following changes have been made:

- When all data up to Day 60 are available, a statistical analysis will be performed. This analysis may lead to the unblinding of some subjects. As a consequence, after Day 60, the study cannot be considered as observer-blind, but will be conducted in a single-blind manner, with subjects' parent(s)/ LAR(s) remaining blinded up to study end (Day ~~365~~***730***). The investigators will not have access to the treatment allocation up to study end (Day ~~365~~***730***).

In **Section 6.4 General study aspects**, the following changes have been made:

- Vaccination will be limited to a maximum of 10 infants per day. In the same investigational center, infants will be vaccinated sequentially, separated by a minimum interval of 60 minutes. ***In addition, to monitor delayed potential reactions following vaccination, there will be a 48-hour observation period following Dose 1 and Dose 2 administration to the first eight infants (corresponding to three to five ChAd155-RSV vaccine recipients) in each Step (1, 2, and 3) before continuing vaccination of more infants (i.e. from the ninth infant). From the ninth infant, infants will be vaccinated sequentially, separated by a minimum interval of 60 minutes.***

In **Section 6.5 Outline of study procedures**, the following changes have been made:

Table 5 List of study procedures

Epoch	**Epoch 001**										**Epoch 002**					
Age	**12-17 mth**										**24-29 mth**	***36-41 mth***				
Type of contact	**Screening**	**Visit 1**	**Visit 2#**	**Visit 3#**	**Visit 4**	**Visit 5**	**Visit 6#**	**Visit 7#**	**Visit 8**	**Visit 9**	**Visit 10**	***Visit 11***	**Unscheduled visit for safety***	**Contact for active/passive surveillance****	**Surveillance for asymptomatic RSV-RTI*****	**Assessment of potential RSV-RTI********
Timepoints	**D-14-*29* to D0**	**D0**	**D1**	**D3**	**D7**	**D30**	**D31**	**D33**	**D37**	**D60**	**D365**	***D730***		**Monthly or Weekly**	**Monthly**	
Informed consent	●															
Informed consent Addendum	●															
Check inclusion/exclusion criteria	●	O														
Collect demographic data	●[a]															
Medical history	●															
Physical examination[b]	●	O	O	O	O	O	O	O	O	O	O	O	O		O	●
Growth monitoring[c]	●					●				●	●	●				
Check contraindications and warnings and precautions		O				O										
Pre-vaccination body temperature		●				●										
Randomization		O														
Vaccine administration		●				●										
Recording of administered treatment number		●				●										
60 minutes post-vaccination observation[d]		●-O				●-O										
Blood sampling for RSV serostatus (1.0 mL)	●															
Blood sampling for assessment of mechanisms of illness (potential ERD; 2.5 mL)																●[l]
Blood sampling for hematology (1.2 mL)	●[e]		●[f]		●[f]	●[f,g]	●[f]		●[f]	●[f]			●[f]			

Epoch	Epoch 001										Epoch 002					
Age	**12-17 mth**										**24-29 mth**	***36-41 mth***				
Type of contact	**Screening**	**Visit 1**	**Visit 2#**	**Visit 3#**	**Visit 4**	**Visit 5**	**Visit 6#**	**Visit 7#**	**Visit 8**	**Visit 9**	**Visit 10**	***Visit 11***	**Unscheduled visit for safety***	**Contact for active/passive surveillance****	**Surveillance for asymptomatic RSV-RTI*****	**Assessment of potential RSV-RTI********
Timepoints	**D-14-*29* to D0**	**D0**	**D1**	**D3**	**D7**	**D30**	**D31**	**D33**	**D37**	**D60**	**D365**	***D730***		**Monthly or Weekly**	**Monthly**	
Blood sampling for biochemistry (1.1 mL)	●[e]				●[f]	●[f,g]			●[f]	●[f]			●[f]			
Blood sampling for CMI response (2.0 mL)	●				●	●[g]			●	●	●					
Blood sampling for humoral response (2.5 mL)	●					●[g]				●	●					
Detection of spontaneous bleeding or easy bruising and evidence of bruising or petechiae			●	●	●		●	●	●							
Examination of limb for petechiae induced by the tourniquet			●		●		●		●							
Surveillance for RSV-RTI, difficulty in breathing and wheezing		O	O	O	O	O	O	O	O	O	O	O		●	●	
Documentation of symptoms and signs of RTI[j]																●
Nasal swab for central testings															●[m]	●[h]
Specimen for local testings																●[n]
Distribution of RTI episode cards[k]		O														
Collection of completed RTI episode cards[k]			O	O	O	O	O	O	O	O	O	O	O		O	O
Transcription of completed RTI episode cards			●	●	●	●	●	●	●	●	●	●	O		●	●
Record any concomitant medications/vaccinations		●	●	●	●	●	●	●	●	●	●	●	●		●	●
Distribution of the subject card		O														
Distribution of diary card		O				O										

Epoch	**Epoch 001**										**Epoch 002**					
Age	**12-17 mth**										**24-29 mth**	***36-41 mth***				
Type of contact	**Screening**	**Visit 1**	**Visit 2#**	**Visit 3#**	**Visit 4**	**Visit 5**	**Visit 6#**	**Visit 7#**	**Visit 8**	**Visit 9**	**Visit 10**	***Visit 11***	**Unscheduled visit for safety***	**Contact for active/passive surveillance****	**Surveillance for asymptomatic RSV-RTI*****	**Assessment of potential RSV-RTI********
Timepoints	**~~D-14~~-*29* to D0**	**D0**	**D1**	**D3**	**D7**	**D30**	**D31**	**D33**	**D37**	**D60**	**D365**	***D730***		**Monthly or Weekly**	**Monthly**	
Return of diary card					O	O			O	O						
Diary card transcription by investigator					●	●			●	●						
Recording of solicited AEs (Day 0-6)		●	●	●		●	●	●					●[i]			
Recording of unsolicited AEs (Day 0-29)		●	●	●	●	●	●	●	●	●			●[i]			
Recording of AE leading to study withdrawal		●	●	●	●	●	●	●	●	●	●	●	●		●	●
Recording of AE of specific interest (RSV-LRTI)		●	●	●	●	●	●	●	●	●	●	●			●	●
Recording of AE of specific interest (spontaneous or excessive bleeding)		●	●	●	●	●	●	●	●	●						
Recording of SAEs		●	●	●	●	●	●	●	●	●	●	●	●		●	●
Recording of SAEs related to study participation, or to a concurrent GSK medication/vaccine	●	●	●	●	●	●	●	●	●	●	●	●	●		●	●
Screening conclusion	●															
Study conclusion											●	●				
Study conclusion for GSK data management										●	●					

Note: the double-bordered lines following Visit 9 (Day 60)***,*** ~~and~~ Visit 10 (Day 365) ***and Visit 11 (Day 730)*** indicate the statistical analyses which will be performed. After Visit 9 (Day 60) the study will be conducted in a single-blind manner, with patients remaining blinded up to study end (Day ~~365~~ ***730***). The investigators will not have access to the treatment allocation up to study end (Day ~~365~~ ***730***).

Visit 2 (Day 1), Visit 3 (Day 3), Visit 6 (Day 31), ~~and~~ Visit 7 (Day 33)***, and Visit 11 (Day 730)*** may take place in the subject's home or at the investigators clinical facility as appropriate to the circumstances in the judgment of the investigator.

$ This is only applicable for subject's parent(s)/LAR(s) who have previously signed the Version 1 of the model ICF

Table 6 Intervals between study visits

Interval	Optimal length of interval	Allowed interval
Screening → Visit 1 (Day 0)	1 - ~~14~~ ***30*** days[1]	-
Visit 1 (Day 0) → Visit 5 (Day 30)	30 days	~~28~~***23*** - ~~35~~***36*** days[2]
Visit 1 (Day 0) → Visit 11 (Day 730)	***730 days***	***700 - 760 days***

[1] Visit 1 should take place no longer than ~~14~~ ***30*** days after the Screening visit. When applicable, a re-screening visit may be scheduled at any time (but only once to assess eligibility; blood for CMI response and humoral response will not be re-sampled). All screening procedures need to be performed within ~~14~~ ***30*** days of Visit 1.

In **Section 6.6.10 Study vaccines administration**, the following change has been made:

- Across each steps, vaccination will be limited to a maximum of 10 infants per day. In the same investigational center, infants will be vaccinated sequentially, separated by a minimum interval of 60 minutes. ***In addition, to monitor delayed potential reactions following vaccination, there will be a 48-hour observation period following Dose 1 and Dose 2 administration to the first eight infants (corresponding to three to five ChAd155-RSV vaccine recipients) in each Step (1, 2, and 3) before continuing vaccination of more infants*** (Refer to Section 9.10.1).

In **Section 6.6.11.2 Blood sampling for hematology and biochemistry**, the following change has been made:

- **at Day 7**, the relevant hematology/biochemistry parameter(s) **must** be re-tested before administration of vaccine Dose 2, either during an unscheduled visit or at the Dose 2 vaccination visit itself (provided results are obtained before vaccination). Only if the concerned parameter(s) is/are within the acceptable range (i.e. within normal range or Grade 1 without clinical relevance), vaccine Dose 2 can be administered. If the concerned parameter(s) is/are not within the acceptable range by Day 35 (which is the maximum allowed interval for the second dosing visit, Visit 5), the infant will not receive Dose 2 but should still continue the study for safety follow-up (up to Day ~~365~~***730***).

In **Section 6.6.14 Surveillance for RSV-RTI and wheezing**, the following changes have been made:

- Surveillance period for RSV-RTI and wheezing will start at Visit 1 (after Dose 1) until the final visit (Visit ~~10~~ ***11*** [Day ~~365~~ ***730***]). In order to timely detect RSV-RTI and to ensure cases are timely captured by the study sites, both active and passive surveillance will be conducted as described in Section 9.2. Contacts for active and passive surveillance will be recorded in the eCRF.
- At the first vaccination visit (Visit 1 [Day 0]), ~~an~~ RTI episode card***s*** will be provided to the subject's parent(s)/LAR(s). The subject's parent(s)/LAR(s) will record the start date and the end date of the following symptoms: cough, runny nose, blocked nose, difficulty in breathing, or wheezing. The subject's parent(s)/LAR(s) will be instructed to return the completed RTI episode card to the investigator at the next visit or by mail (e-mail or postal mail). The investigator will transcribe the collected information into the eCRF. Any unreturned RTI episode card will be sought from the subject's parent(s)/LAR(s) through telephone call(s) or any other convenient procedure.

In **Section 6.6.18 Study conclusion**, the following changes have been made:

- At the last visit (Visit ~~10~~ ***11*** [Day ~~365~~ ***730***]), the investigator will:

In **Section 6.7.2 Biological samples**, the following changes have been made:

Table 7 Biological samples

	Total volume of blood collected for each subject from Screening to Visit ~~10~~ *11* (Day ~~365~~*730*)*	**36.9 mL**

In **Section 6.7.4.1 Immunological read-outs**, the following changes have been made:

Table 12 Immunological read-outs

Blood sampling timepoint			No. subjects	Component	Components priority rank
Type of contact and timepoint	Sampling timepoint	System			
Screening (Day -~~14~~*29* to Day 0)	Pre-Vaccination	Whole blood	~96	IL-2, CD40L, 41BB, TNF-α, IFN-γ, IL-13, IL-17 secreting T-cells	1
		Serum	≥ 96	Respiratory Syncytial virus Ab.IgG (RSV F or G antibody)	1
			~96	Anti-RSV A Neutralizing Antibody	2
				Respiratory Syncytial Virus F protein Ab.IgG (Anti-RSV PreF antibody)	3
				Respiratory Syncytial Virus F protein Ab.IgG (Palivizumab competing Ab)	4

In **Section 7.1 Description of study vaccines**, the following changes have been made:

Table 14 Study vaccines

Placebo 0.5	Formulation buffer S9b	Na_2HPO_4=1.3mg; KH_2PO_4=186µg; NaCl=3.85mg; KCl=100µg; $MgCl_2$=50µg *	Clear liquid	0.5 ml	2
Placebo 0.15	Formulation buffer S9b	Na_2HPO_4=0.4mg; KH_2PO_4=56µg; NaCl=1,16mg; KCl=30µg; $MgCl_2$=15µg *	Clear liquid	0.15 ml	2

In **Section 7.6.1 Recording of concomitant medications/products and concomitant vaccinations**, the following change has been made:

- Any concomitant vaccination administered in the period starting 30 days before the first dose of study vaccines and ending at the last study visit (Day -29 to Day ~~365~~*730*).

In **Section 9.2 Surveillance for RSV-RTI, difficulty in breathing and wheezing episodes**, the following changes have been made:

- Each subject will be under surveillance for RSV-RTI, difficulty in breathing and wheezing episodes from the administration of Dose 1 (Visit 1) and will continue until the final visit (Visit ~~10~~ *11*). Surveillance will be performed via phone or e-mail contacts and assessment visits (refer to Figure 3). The passive and active surveillance contacts can also be made by/with the person designated by the parent(s)/LAR(s) (e.g. grandparents, nanny) as long as the parent(s)/LAR(s) have given approval.

Figure 3 Decision tree for passive and active surveillance contacts

[6] In case of worsening of symptoms, optional sample if the previous ~~swab~~ ***specimen*** was RSV-positive.

In **Section 9.2.3 Active surveillance**, the following changes have been made:

- There will be contact between the investigator/study staff and subject's parent(s)/LAR(s) on a regular basis (weekly during the RSV season and every month outside the RSV season). If there has not been a contact through a clinic visit (i.e. Visits 1, 2, 3, 4, 5, 6, 7, 8, 9, 10, ***11***), a passive surveillance contact (refer to Section 9.2.1), an assessment visit (refer to Section 9.2.4) or a surveillance for asymptomatic RSV infection (refer to Section 9.2.2), then the investigator/study staff will contact the subject's parent(s)/LAR(s). The active surveillance will be performed by phone, mobile phone or e-mail. During each active follow-up contact, the investigator/study staff will:

In S**ection 9.3.1 Time period for detecting and recording adverse events, adverse events of specific interest and serious adverse events**, the following changes have been made:

Table 18 Reporting periods for collecting safety information

Visit	SCR	V1	V2	V3		V4		V5	V6	V7		V8		V9	V10	*V11*
Days	~~-14~~*29* to 0	0	1	3	6	7	29	30	31	33	36	37	59	60	365	*730*
AEs/SAEs leading to withdrawal from the study																
AE of specific interest (RSV-LRTI)																
SAEs																
SAEs related to study participation (start at signature of informed consent form) or concurrent GSK medication/vaccine																

In **Section 9.10.1 Limited vaccination**, the following change has been made:

- In the same investigational center, all infants should be vaccinated sequentially and at least 60 minutes apart to allow monitoring of any acute events (e.g. hypersensitivity reaction). All infants should be closely observed (visual follow-up as well as measurement of vital signs) for at least 60 minutes after vaccination. Vital signs are body temperature, HR, and RR. Vital signs are preferably measured when the infant is calm. ***In addition, to monitor delayed potential reactions following vaccination, there will be a 48-hour observation period following Dose 1 and***

Dose 2 administration to the first eight infants (corresponding to three to five ChAd155-RSV vaccine recipients) in each Step (1, 2, and 3) before continuing vaccination of more infants (i.e. from the ninth infant). From the ninth infant, infants will be vaccinated sequentially, separated by a minimum interval of 60 minutes. Refer to the SPM for more details about the 48-hour observation period after the first eight subjects vaccinated in each Step.

In **Section 9.10.2 Internal safety review committee (iSRC) oversight**, the following change has been made:

- LRTI associated with RSV infection (AE of specific interest) occurring from Day 0 to Day ~~365~~***730***.

In **Section 9.10.3 Independent Data Monitoring Committee (IDMC) oversight**, the following changes have been made:

- The IDMC will receive the following safety data within 48 hours upon GSK becoming aware of:
  - Fatal SAEs occurring from Day 0 to Day ~~365~~***730***.
  - Life-threatening SAEs occurring from Day 0 to Day ~~365~~***730***.
  - Related SAEs occurring from Day 0 to Day ~~365~~***730***.
  - LRTI associated with RSV infection (AE of specific interest) occurring from Day 0 to Day ~~365~~***730***.
- The IDMC will receive the following safety data ***approximately*** monthly from beginning of ***the first*** RSV season ***and the frequency will be adapted based on the instruction of the IDMC***:
  - Cumulative reports of the incidence of RSV-RTI, RSV-LRTI and RSV-severe LRTI. ~~The frequency will be initially monthly from the beginning of the RSV season and will be adapted based on the instruction of the IDMC~~.

In **Section 9.10.5 Holding rules**, the following change has been made:

Table 21 Holding rules assessed by the investigator

Holding Rule	Event	Number of infants/group		
		Step 1	Step 2	Step 3
1c	Any local or general solicited AE leading to hospitalization ***that cannot reasonably be attributed to a cause other than vaccination.***	≥ 1	≥ 1	≥ 1

In **Section 11.2 Secondary endpoints**, the following changes have been made:

- Occurrence of SAEs from study start (Day 0) up to study conclusion (Day ~~365~~***730***) in all subjects.

- Occurrence of RSV-LRTI (AE of specific interest) as from Dose 1 administration up to study conclusion (Day ~~365~~***730***) in all subjects.
- Occurrence of RSV-RTI, RSV-LRTI, severe RSV-LRTI (according to case definitions) as from Dose 1 administration up to study conclusion (Day ~~365~~***730***) in all subjects.

In **Section 11.5.2 According-to-protocol cohort for analysis of immunogenicity**, the following changes have been made:

- Who received ~~two doses~~ ***at least one dose*** of study vaccine.

In **Section 11.6.4 RTI and LRTI**, the following changes have been made:

- For the analysis of RTI and LRTI, all cases will be definitively classified as either RTI, LRTI or severe LRTI according to the case definitions presented in Table 4, and the association to RSV infection will be assessed by quantitative PCR as primary analysis. ***The analysis may also be done using the available WHO case definitions for RTI, LRTI and severe LRTI.***

In **Section 11.10 Analysis of RTI and LRTI**, the following change has been made:

- The analysis will be performed on the TVC on two pooled groups (i.e. pooled RSV and pooled Placebo) ***separately for first and second RSV seasons and overall.***
- As primary analysis, the assessment of RSV infection will be performed using the quantitative PCR according to case definitions presented in Table 4. ***The analysis may also be done using the available WHO case definitions for RTI, LRTI and severe LRTI.***

In **Section 11.12.1 Sequence of analyses**, the following changes have been made:

- The statistical analyses will be performed in ~~2~~ ***3*** steps:
  - A first analysis will be performed when all data up to Day 60 (i.e. data that are as clean as possible) are available. Additional safety data available at the time of this analysis will be described. At this point, the statistician will be unblinded (i.e. individual subject treatment assignments will be available) and the study will be conducted in a single-blind manner, with patients remaining blinded up to study end (Day ~~365~~***730***). Summary results may unblind some specific subjects but no individual listings will be provided and the investigators will not have access to the treatment allocation up to study end (Day ~~365~~***730***)***, except in case of emergency unblinding (see Section 9.8)***.
  - ***An analysis will be performed when all data up to Visit 10 (Day 365; i.e. data that are as clean as possible) are available. No individual listings will be provided.***
  - The final analysis will be performed when all data up to study conclusion (Day ~~365~~***730***) are available. An integrated clinical study report containing all data will be written and made available to the investigators at that time.

In **Appendix A "Laboratory Assays"**, the description of the different assays performed during the study has been updated in order to be consistent throughout the different RSV project documents.

Intracellular staining (ICS)

Within 4 hours post phlebotomy, approximately 350 µL of undiluted whole blood are incubated at 37°C with the relevant antigen (in this case pools of 15-mer peptides overlapping by 11 amino acids and spanning the sequences of F, N, and M2-1 proteins), negative control (medium), or positive control (staphylococcal enterotoxin B) for 2 hours in the presence of anti-CD28 and anti-CD49d antibodies. Brefeldin A, a protein secretion inhibitor, is then added and the mix is incubated overnight. Red cells are then lysed and white cells washed, fixed, cryopreserved, and stored until analysis by cytometry.

Once ready for cytometry analysis, white cells are thawed in batches, washed, and incubated with fluorochrome-conjugated antibodies specific to CD4 and CD8 surface markers. Then, cells are permeabilized and stained with fluorochrome-conjugated antibodies specific for the following immune markers: ~~For pediatric blood samples, undiluted whole blood is stimulated at 37°C with the relevant antigen – in this case peptide pools covering proteins F, N and M2-1 – for 2 hours in the presence of anti-CD28 and anti-CD49d antibodies. An inhibitor of intracellular protein transport (Brefeldin A) is added and cells are incubated overnight. Red cells are then lysed and white cells washed, fixed, cryopreserved, and stored until cytometric analysis.~~

~~Cells are subsequently thawed in batches, washed, and incubated with fluorochrome-conjugated antibodies specific to CD3, CD4 and CD8 surface markers. Cells are then permeabilized and stained with fluorochrome-conjugated antibodies specific for immune markers such as~~ IL-2, IFN-γ, TNF-α, CD40L, IL-13, IL-17, and 41BB. ~~After washing, flow cytometric acquisition is performed and the results are expressed as the frequency of CD4+ and CD8+ T-cells expressing any cytokine or a combination of cytokines.~~

~~Other immune markers could be added to characterize further the antigen-specific CD4+ or CD8+ T-cells.~~

After washing, acquisition of data is performed via a flow cytometer.

The results are expressed as the frequency of CD4 or CD8 T-cells expressing, per million of CD4 or CD8 T-cells:

- ***At least one immune marker (to detect and measure the CD4 or CD8 T-cell response).***
- ***Any Th specific immune marker (to determine the Th profile of the CD4 response).***

Neutralization assay

~~The assay comprises several steps. First, a virus neutralization step is performed by incubating fixed quantity of live RSV virus (RSV A long strain) with serial dilutions of test serum. The serum-virus reaction mixture is then transferred onto a monolayer of~~

~~Vero cells (African Green Monkey, kidney, *Cercopitheus aethiops*) and incubated for 3 days. Virus-infected cells are detected by an immunofluorescence assay, using a two-step detection system with a primary goat anti-RSV antibody and a secondary fluorochrome-tagged antibody (anti-Goat IgG-FITC). This antibody system detects host cell-associated RSV antigens and allows visualization of plaques appearing on the cell monolayer by immunofluorescence. The viral plaques are counted as plaque-forming units using an automated microscope reader.~~

~~The serum neutralizing antibody titer is reported as the inverse of the serum dilution which yields a 60% reduction in the number of viral plaques compared to the virus control without serum (Effective Dose: ED60)~~ ***First, virus neutralization is performed by incubating a fixed amount of RSV-A long strain (ATCC No. VR-26) with serial dilutions of the test serum. Then, the serum-virus mixture is transferred onto a monolayer of Vero cells (African Green Monkey, kidney, Cercopitheus aethiops, ATCC CCL-81) and incubated for three days to allow infection of Vero cells by non-neutralized viruses and the formation of plaques in the cell monolayer. Following the fixation period, RSV-infected cells are detected using a primary antibody directed against RSV (anti-RSV IgG) and a secondary antibody conjugated with fluorescein isothiocyanate, allowing the visualization of plaques by immunofluorescence. Viral plaques are counted using an automated microscope coupled to an image analyzer (Scanlab system with Axiovision software). For each serum dilution, a ratio, expressed as a percentage, is calculated between the number of plaques at that dilution and the number of plaques in the virus control wells (no serum added). The serum neutralizing antibody titer is expressed in ED60 (Estimated Dilution 60) and corresponds to the inverse of the interpolated serum dilution that yields a 60% reduction in the number of plaques compared to the virus control wells*** as described by others [Barbas, 1992; Bates, 2014].

ELISA

- Anti-RSV protein F ELISA

  ~~The anti-RSV protein F ELISA is an indirect ELISA allowing the detection and the quantification of specific IgG antibodies directed against the RSV F protein in human serum samples.~~

  ~~In summary, protein F antigen purified from Chinese Hamster Ovary expression system is adsorbed onto a 96-well polystyrene microplate. After a washing and a blocking step, dilutions of serum samples, controls and standards are added to the coated microplate. After incubation, the microplate is washed to remove unbound primary antibodies. Bound IgGs are detected by the addition of a goat anti-human IgG antibody, conjugated to horse-radish peroxidase. Bound antibodies are quantified by the addition of the horse-radish peroxidase substrate, tetramethylbenzidine and hydrogen peroxide, whereby a colored product develops proportionally to the amount of anti-RSV PreF IgG antibodies present in the serum sample. The antibody concentration is expressed in arbitrary EU/mL.~~

  The anti-F protein IgG ELISA is an indirect ELISA allowing the detection and the quantitation of specific IgG antibodies directed against the RSV F protein in human serum samples.

First, F protein antigens purified from CHO expression system are coated onto 96-well microplates. Then, after a washing and a blocking step, serial two-fold dilutions of test sera, controls, and reference standard are incubated to allow specific binding of antibodies directed against the F protein antigens. Bound IgG are detected by addition of a goat anti-human IgG antibody conjugated to horseradish peroxidase. After a washing step, the horseradish peroxidase substrate solution (TMB/H_2O_2) is added and a colored product develops proportionally to the amount of anti-F protein IgG antibodies present in the test serum. The color is quantified by reading the optical densities at 450-620 nm using a spectrophotometer. Antibody concentrations of individual serum and control samples are determined after interpolation from the reference standard curve using a four-parameter equation and are expressed in arbitrary ELISA units (EU)/mL.

- Palivizumab competitive assay

  Palivizumab monoclonal antibody (*Synagis*) is used as a passive treatment that protects against RSV infection by binding to the antigenic site II epitope of the RSV F antigen. ~~The palivizumab competitive assay is based on the competitive binding process between the labeled antibody (biotinylated palivizumab) and non-labeled antibody (palivizumab-equivalent antibodies in serum used as reference) targeting the same epitope on a coated antigen. The results of the palivizumab competitive assay ELISA are expressed as palivizumab equivalent antibodies in µg/mL.~~

  First, F protein antigens purified from CHO expression system are coated onto 96-well microplates. Then, after a washing and a blocking step, serial two-fold dilutions of test sera, positive control serum, and palivizumab antibody reference standard are added in sequence with competitor antibodies (horseradish peroxidase-conjugated palivizumab) and incubated to allow specific binding of antibodies directed against the F protein antigens. If palivizumab-like antibodies are present in serum samples, they will compete with the horseradish peroxidase-conjugated palivizumab antibodies for binding to the F protein coated antigen. After a washing step, the horseradish peroxidase substrate solution (TMB/H_2O_2) is added and a colored product develops in a manner that is inversely proportional to the amount of palivizumab-like antibodies contained in the test serum. The color is quantified by reading the optical densities at 450-620 nm using a spectrophotometer. Antibody concentrations of individual serum and control samples are determined after interpolation from the ELISA standard curve using a four-parameter equation and are expressed as palivizumab-equivalent antibodies in microgram per millilitre (µg/mL).

PCR

- Quantitative PCR able to discriminate RSV-A and RSV-B subtypes:

  ~~RSV-A and RSV-B RNAs, extracted from the nasal swab are detected in a duplex format using specific amplification primers and fluorescent TaqMan™ probes. The process involves viral RNA extraction, conversion of RNA to complementary deoxyribonucleic acid by reverse transcription and detection by real- time PCR reaction using a calibration curve (absolute quantification)~~.

Briefly, RSV A and RSV B RNAs extracted from the nasal swabs are detected in a duplex PCR format using specific amplification primers and fluorescent probes designed in the RSV N gene, encoding the RSV nucleocapsid protein. The process involves nucleic acids extraction, conversion of RNA to complementary deoxyribonucleic acid by reverse transcription and detection by real- time PCR reaction using a calibration curve (absolute quantitation). The RSV viral load is reported as copies of RSV RNA per mL of sample.

- Qualitative multiplex PCR for detection of a panel of 17 viruses: xTAG™ RVP FAST assay:

  ~~xTAG™ RVP FAST assay is a qualitative nucleic acid multiplex tests intended for the simultaneous detection and identification of multiple respiratory virus nucleic acids in nasal/nasopharyngeal swabs from individuals suspected of respiratory tract infections. After nucleic acid extraction, a multiplexed PCR reaction is performed to amplify the regions of interest in the targeted infectious agent genes. The PCR reaction product is then subjected to a hybridization/detection step and attached to an xTAG universal tag sequence. The 5' universal tag sequence is hybridized to the complementary anti-tag sequence coupled to a particular bead set. The hybridized beads are detected and results are analyzed by the data analysis software.~~

  After extraction of nucleic acids from the sample, a single multiplex RT-PCR is performed using random primers. Then, each complementary deoxyribonucleic acid is detected by virus-specific primers, each primer containing a unique tag sequence. A deoxyribonucleic acid polymerase extends the primer when there is a perfect match on the 3' end of the virus specific primer. Biotin-dCTP is incorporated into the extending chain if extension occurs.

  Tagged amplified deoxyribonucleic acid is mixed with Luminex beads. These beads are coated with anti-tag oligonucleotides complementary to the tag sequences included in the virus-specific primers. These beads are colored making them spectrally distinguishable from each other. Each tagged primer hybridizes only to its unique anti-tag complement; therefore, each bead color represents a specific virus through the bead/anti-tag/tagged primer association.

  Revelation is performed by using a fluorescent reporter molecule (streptavidin-phycoerythrin) that binds to the Biotin-dCTP included in the extended sequences. The beads are then analyzed by the Luminex® 200 instrument which corresponds to a flow-cytometer containing two lasers: one laser allows identification of the color-coded bead associated to each virus and one laser allows identification of the presence or absence of extended primer through the reporter molecule. The data generated by the Luminex® 200 system are expressed in Median Fluorescence Intensity units per bead, which are translated in a positive or negative status for each target.

GlaxoSmithKline Biologicals SA

Vaccines R &D

Protocol Amendment 2

eTrack study number and Abbreviated Title:	204838 (RSV PED-002)
IND number:	16999
EudraCT number:	2016-000117-76
Amendment number:	Amendment 2
Amendment date:	08 June 2017
Co-ordinating author:	PPD (Scientific Writer, Keyrus Biopharma contractor for GSK Biologicals)

Rationale/background for changes:

- During initial implementation of this study, parents have given feedback that the number of visits and blood tests is challenging to accommodate with busy family schedules and is demanding for young children. This amendment reduces the burden on families whilst maintaining the intended close oversight of the potential risk of thrombocytopenia, by reducing the number and frequency of required blood tests and visits unless clinically required. Assessment of platelet counts (currently at Days 1, 7, 31 and 37) will now be repeated at Day 7 and/or Day 37 in those infants with a Grade 1 or greater abnormality detected at either Day 1 and/or Day 31. This is because the nadir of the platelet count occurred on the first day following dosing in the preclinical data. This modification of monitoring is judged by the Independent Data Monitoring Committee (IDMC) and GSK to be appropriate, as the sampling plan captures any potential initial fall and then subsequent recovery. The clinic visits at Day 3 and Day 33 including a clinical examination of the child for evidence of petechiae may be replaced by a phone call to inquire of the infants' general health and to solicit reports of a rash, spontaneous bleeding or easy bruising, which are all indicative of thrombocytopenia. As an additional safety measure the subjects' parent(s)/legally acceptable representative(s) (LAR[s]) will be instructed to contact the investigator/study staff immediately if their child develops a rash, in order to detect any thrombocytopenic petechiae or purpura. This will supplement the current instruction to report spontaneous bleeding or easy bruising in the month following vaccination. As there is no absolute requirement for sampling for hematology on Day 7 or 37, the other tests of biochemistry and cell mediated immunity (CMI) are not requested under this amendment at these timepoints.
- It has been clarified that blood samples for CMI and for humoral immunogenicity may be postponed until after the RSV serostatus is known and eligibility is confirmed up to or on Day 0 (but before vaccination).
- The age range for enrolment has been expanded an additional 6 months, from 12 to 17 months initially, to 12 to 23 months. These children are still at risk of respiratory syncytial virus (RSV) disease and could potentially benefit from this vaccine. This would also increase the number of subjects screened who are likely to be previously

exposed to RSV. Increasing the pool of RSV-seropositive children will decrease the number of failures to enroll after screening.

- The following inclusion criterion has been removed, to be able to include those infants who despite being mildly underweight or premature have had normal subsequent courses: "Born full-term (i.e. after a gestation period of 37 to less than 42 completed weeks) with a minimum birth weight of 2.5 kg". Other criteria pertaining to current weight, health status of the child, and *Synagis* administration will exclude infants who have experienced prematurity and its associated complications.
- Because enrolment in the trial is slower than anticipated, the safety monitoring oversight plan of the IDMC is adapted to provide regular review of accumulating safety data every four weeks in addition to the currently planned review at end of each dose level. Moving to a next dose level will be done in the absence of a safety concern detected by the IDMC on the four weekly reports and after administration of the vaccine to all 32 subjects in the previous Step. It will be ensured that, as a minimum, the IDMC will have reviewed safety data of at least 16 subjects for two doses and given their formal approval prior to enrolling subjects to the next dose level.
- To allow the extension of the recruitment network, some investigational sites without a laboratory in proximity capable to perform the whole blood stimulation (WBS) necessary for the CMI assay will be allowed to participate. Therefore, blood sampling for the assessment of CMI will only be performed for subjects recruited in sites with a WBS capable laboratory in proximity.

The following minor changes and clarifications have been made:

- Further clarification is made on which visits may take place in the subject's home as appropriate to the circumstances and in the judgment of the investigator (not allowed if blood sampling for immune response and/or vaccine administration).
- The priority ranking in blood sampling is amended to hematology > biochemistry > humoral response > CMI response. This is primarily a study of safety and so venipuncture will not be repeated for CMI or humoral response in case of insufficient volume at any timepoint, if safety samples have been secured.
- Safety data from study 201974 (RSV PED-001) in healthy adults have been added.
- It has been clarified that RSV infections will be assessed using standardized case definitions based on the available World Health Organization (WHO) case definitions [Modjarrad, 2016].
- Clarification on the reporting of temperature excursions has been added. Temperature excursion above -60°C and below -90°C for the ChAd155-RSV vaccine should be reported.
- The endpoints related to CMI evaluation have been clarified.
- The kit used for multiplex respiratory viral panel testing has been changed.
- In addition, some typos have been corrected and other administrative changes have been made throughout the protocol in order to add clarity.

- Finally, changes in study personnel have been included on the protocol cover page.

Amended text has been included in *bold italics* and deleted text in ~~strikethrough~~ in the following sections:

Throughout the document, the following changes have been made:

- The age range of infants to be enrolled has been widened, from 12 to 17 months initially, to 12 to 23 months.

On **the protocol cover page**, the following changes have been made:

Contributing authors

- PPD (***Expert*** Scientific Writer~~, Keyrus Biopharma contractor for GSK Biologicals~~)
- PPD ***(Scientific Writer, Keyrus Biopharma contractor for GSK Biologicals)***
- PPD ***(Lead Statistician)***
- PPD (~~Vaccine~~ ***Clinical Trial*** Supply ***Manager*** ~~Coordinator~~, Aixial contractor for GSK Biologicals)
- PPD ***(Oversight Data Manager, Keyrus Biopharma contractor for GSK Biologicals)***

In the **Synopsis**, the following changes have been made:

Rationale for the study and study design

Rationale for the study design

- **Study population**: The immunogenicity, safety and reactogenicity of the pediatric candidate RSV vaccine (ChAd155-RSV vaccine) ~~are currently being~~ ***has been*** evaluated in healthy adult subjects aged 18 to 45 years (study 201974 [RSV PED-001]).
- **Staggered design with 3 steps:**

  ~~Within each step, a~~***A***n internal Safety Review Committee (iSRC) will review ***all accumulating*** ~~accumulated~~ safety data three weeks after the start of vaccination and then about every three weeks until the IDMC ~~reviews the data~~ ***has reviewed all safety data up to 30 days after administration of Dose 2 in Step 3 (i.e. Day 60)***.

  The IDMC will review accumulating reactogenicity and safety data monthly throughout the period of vaccination (up to 30 days after administration of Dose 2 in Step 3, i.e. Day 60). The IDMC will also review incidence of all serious

adverse events (SAEs) and incidence of RSV-respiratory tract infections (RTI), RSV-LRTI, RSV-severe LRTI and RSV-RTI leading to hospitalization monthly after the start of vaccination and from beginning of the first RSV season to the end of the study.

Both iSRC and IDMC will receive critical safety data (i.e. SAEs and adverse events of specific interest) within 48 hours upon GSK becoming aware of it.

The iSRC and IDMC will determine whether any of the predefined study holding rules are met. If this is the case, vaccination in the study will be immediately put on hold. At any time, the IDMC may recommend to the sponsor to suspend the enrolment to the study and/or vaccination based on the safety data regularly arising in this study.

~~Moving to a next step will be conditional to the favorable outcome of the evaluation of safety data obtained up to 7 days after administration of the second vaccine dose in the previous step (for all subjects), performed by an IDMC.~~ ***Dose escalation will proceed after administration of the vaccine to all 32 subjects in the previous Step, and in the absence of a safety concern detected by the IDMC in the regular monitoring of accumulating safety data. It will be ensured that, as a minimum, the IDMC will have reviewed safety data of at least 16 subjects for two doses and given their formal approval prior to enrolling subjects to the next dose level.***

Objectives

Tertiary

- To further evaluate the CMI ***profile*** induced by two IM doses of three dose levels of the RSV investigational vaccine when administered according to a 0, 1-month schedule, up to Day 365 in RSV-seropositive infants aged 12 to ~~17~~***23*** months***.***

Study design

- End of Study: Last testing results released of samples collected at Visit 11 (Day 730)****.***

  **** Up to Visit 11 (Day 730), there will be monthly nasal swab to detect asymptomatic RSV infections during the RSV season and if following active or a passive surveillance contacts, a subject presents symptoms of RTI, a nasal swab will be collected.***

- Sampling schedule:

  – Blood samples for **biochemistry** will be taken from all infants at Screening (29 days before first vaccination to Day 0) and on ~~Day 7,~~ Day 30~~, Day 37,~~ and Day 60. Blood samples for **hematology** will be taken from all infants at Screening (29 days before first vaccination to

Day 0) and on Day 1, ~~Day 7,~~ Day 30, Day 31, ~~Day 37,~~ and Day 60. ***Day 7 hematology testing will be performed if any ≥ Grade 1 for platelet decrease is detected on Day 1 after vaccination, to ensure its resolution. Similarly, hematology testing on Day 37 will be performed if any ≥ Grade 1 for platelet decrease is detected on Day 31***. A clinical ~~history and~~ examination with special attention given to detection of spontaneous bleeding or easy bruising and evidence of bruising or petechiae will be performed ***by the investigator/study staff*** on Day 1, Day 3 and Day 7 after each vaccination (i.e. Day 1, Day 3, Day 7, Day 31, Day 33, and Day 37). ***The visits on Day 3 and Day 33 may be replaced by a phone call by the investigator/study staff to inquire of the infants' general health and to solicit reports of a rash, spontaneous bleeding or easy bruising.*** On Day 1, ~~Day 7,~~ ***and*** Day 31, ~~and Day 37,~~ special attention will also be given to the detection of petechiae or evidence of petechiae induced by the tourniquet. ***This special attention to detection of petechiae or evidence of petechiae induced by the tourniquet will also be given on Day 7 and/or Day 37 in case hematology testing will be performed on that day***. Further testing may be required to investigate a finding or guide subject management based on the investigator***'s*** clinical judgment. Subjects' parent(s)/LAR(s) will be instructed to contact the investigator/study staff if their child presents symptoms of spontaneous bleeding or easy bruising ***or if their child develops a rash in the month*** following vaccination***, in order to detect any thrombocytopenic petechiae or purpura. The investigator will, based on his/her medical judgement, appropriately investigate infants with clinical suspicion of low platelets.***

- Blood samples for **CMI** ***are limited to those in investigational sites with a laboratory in proximity capable to perform WBS necessary for the CMI assay. In those sites, samples*** will be taken from all subjects at Screening ~~***(or alternatively during an additional visit before Day 0)***~~ and on ~~Day 7,~~ Day 30, ~~Day 37,~~ Day 60, and Day 365***.***
- Blood samples for **humoral immunogenicity** will be taken from all subjects at Screening ~~***(or alternatively during an additional visit before Day 0)***~~ and on Day 30, Day 60, and Day 365.

- ***Study visits: Visit 1 (Day 0), Visit 5 (Day 30), Visit 9 (Day 60) and Visit 10 (Day 365) must be performed at the***

investigators clinical facility. Visit 2 (Day 1), Visit 4 (Day 7), Visit 6 (Day 31), Visit 8 (Day 37) and Visit 11 (Day 730) (no blood sampling for immune response and no vaccine administration) may take place in the subject's home or at the investigators clinical facility as appropriate to the circumstances in the judgment of the investigator. Visit 3 (Day 3) and Visit 7 (Day 33) may take place at the investigators clinical facility or the investigator/clinical staff may call the subjects' parent(s)/LAR(s), as appropriate to the circumstances in the judgment of the investigator.

Case definition

During the analysis of the study, all cases identified during the surveillance of RSV-RTI will be definitively classified as either RTI, LRTI, ~~and~~ severe LRTI ***or very severe LRTI*** according to the ***standardized*** case definitions ~~described in~~ ***(see*** Synopsis Table 4***) based on the available World Health Organization (WHO) case definitions***. ~~The analysis may also be done using the available World Health Organization (WHO) case definitions for RTI, LRTI and severe LRTI.~~

Synopsis Table 4 Case definitions for data analysis

RSV-RTI	RSV-LRTI	severe RSV-LRTI
RSV A or B positive by quantitative PCR AND Runny nose, OR Blocked nose, OR Cough	RSV A or B positive by quantitative PCR AND Infant with RTI AND SpO_2 < 95% (measured in room air by pulse oximetry), OR RR increase: > 40/minutes 12 - 30 months of age	RSV A or B positive by quantitative PCR AND Infant with LRTI AND SpO_2 < 92% (measured in room air by pulse oximetry), OR Difficulty breathing leading to: Irritability/agitation, OR Lethargy/sleepiness, OR Lower chest wall indrawing, OR Reduced/no vocalization, OR Apnea > 20 seconds, OR Cyanosis, OR Stop feeding well/dehydration
All-cause RTI	**All-cause LRTI**	**All-cause severe LRTI**
Runny nose, OR Blocked nose, OR Cough	Infant with RTI AND SpO_2 < 95% (measured in room air by pulse oximetry), OR RR increase: > 40/minutes 12 - 30 months of age	Infant with LRTI AND SpO_2 < 92% (measured in room air by pulse oximetry), OR Difficulty breathing leading to: Irritability/agitation, OR Lethargy/sleepiness, OR Lower chest wall indrawing, OR Reduced/no vocalization, OR Apnea > 20 seconds, OR Cyanosis, OR Stop feeding well/dehydration

PCR: polymerase chain reaction; **RR**: respiratory rate; **SpO_2**: blood oxygen saturation; **RTI**: respiratory tract infections; **LRTI**: lower respiratory tract infections

RSV-RTI	***Runny nose OR blocked nose OR cough*** ***AND*** ***Confirmed RSV infection [4]***
RSV-LRTI	***History of cough OR difficulty breathing [1]*** ***AND*** ***SpO_2 < 95% [2], OR RR increase [3]*** ***AND*** ***Confirmed RSV infection [4]***
RSV-severe LRTI	***Meeting the case definition of RSV-LRTI*** ***AND*** ***SpO_2 < 93% [2], OR lower chest wall in-drawing***

RSV-very severe LRTI	***Meeting the case definition of RSV-LRTI*** ***AND*** ***SpO_2 < 90% [2], OR inability to feed, OR failure to respond / unconscious***
RSV hospitalization	***Confirmed RSV infection [5]*** ***AND*** ***Hospitalized for acute medical condition [6]***
All-cause LRTI	***History of cough OR difficulty breathing [1]*** ***AND*** ***SpO_2 < 95% [2], OR RR increase [3]***

Definitions based on [Modjarrad, 2016]
LRTI = lower respiratory tract infections; RR = respiratory rate; RTI = respiratory tract infections; SpO_2 = blood oxygen saturation.
[1] Based on history reported by parents/LARs and includes difficulty breathing (e.g. showing signs of wheezing or stridor, tachypnoea, flaring [of nostrils], chest in-drawing, apnoea) associated with nasal obstruction.
[2] For blood oxygen saturation (SpO_2), the lowest value monitored will be used.
[3] RR increase defined as ≥ 40/minute (12 months of age or above).
[4] RSV infection confirmed on nasal swab positive for RSV A or B by quantitative Reverse Transcription Polymerase Chain Reaction (qRT-PCR).
[5] RSV sampling and testing is based on medical judgment of medical practitioner or driven by algorithm.
[6] Hospitalization is defined as a medical decision that the infant requires admission for observation or treatment.

Endpoints

Primary

- Occurrence of AEs from first vaccination (Day 0) up to Day 60~~, in all infants~~.
  - Occurrence of episode of spontaneous or excessive bleeding (AE of specific interest), during a 30-day follow-up period after each vaccination~~, in all subjects~~.
  - Occurrence of any biochemical (alanine aminotransferase, aspartate aminotransferase and creatinine) laboratory abnormalities at Screening, ~~Day 7~~, Day 30,~~ Day 37,~~ and Day 60.

Secondary

- Occurrence of SAEs from study start (Day 0) up to study conclusion (Day 730) ~~in all subjects~~.
- Occurrence of RSV-LRTI (AE of specific interest) as from Dose 1 administration up to study conclusion (Day 730) ~~in all subjects~~.
- Occurrence of RSV-RTI, RSV-LRTI, severe RSV-LRTI (according to ***standardized*** case definitions) as from Dose 1 administration up to study conclusion (Day 730) ~~in all subjects~~.

- Magnitude of the CMI response to the investigational RSV vaccine, pre-vaccination (Screening), post-Dose 1 (~~Day 7 and~~ Day 30) and post-Dose 2 (~~Day 37,~~ Day 60 and Day 365)~~, in all subjects~~**.**
  - CD3+/CD4+ ~~and CD3+/CD8+~~ T-cells expressing ~~at least one marker~~ ***at least two markers among CD40L, IL-2, TNF-α, and IFN-γ upon stimulation with F, N and M2-1 peptide pools***.
- Humoral response to the investigational RSV vaccine, pre-vaccination (Screening), post-Dose 1 (Day 30) and post-Dose 2 (Day 60 and Day 365)~~, in all subjects~~:
- Humoral response to the investigational RSV vaccine, pre-vaccination (Screening), post-Dose 1 (Day 30) and post-Dose 2 (Day 60)~~, in all subjects~~:

Tertiary

- CMI response ***profile (Th1, Th2, Th17)*** to the investigational RSV vaccine, pre-vaccination (Screening), post-Dose 1 (~~Day 7 and~~ Day 30) and post-Dose 2 (~~Day 37,~~ Day 60 and Day 365)~~, in all subjects~~**.**
  - CD3+/CD4+ and***/or*** CD3+/CD8+ T-cells expressing ~~any marker or a combination of markers, for determination of Th profile~~ ***at least one or any combination of immune marker(s) among CD40L, 41BB, IL-2, TNF-α, IFN-γ, IL-13 and IL-17 upon stimulation with F, N and M2-1 peptide pools***.
- Any further exploratory immunology to detect ***disease-related or*** vaccine-related immune responses, such as but not limited to:

In the **List of abbreviations**, the following changes have been made:

WBS: ***Whole blood stimulation***

In Section Trademarks, the following changes have been made:

Trademarks of the GSK group of companies	***Generic description***
Bexsero	***Meningococcal group B vaccine (recombinant, adsorbed)***

Trademarks not owned by the ~~GlaxoSmithKline~~ *GSK* group of companies	**Generic description**
Synagis~~™~~ (MedImmune)	Recombinant humanized monoclonal anti-RSV antibodies
Allplex (Seegene)	***Respiratory panel assay***

In Section 1.1.6 Pre-clinical *and clinical* experience, the following changes have been made:

The immunogenicity, safety and reactogenicity of the pediatric candidate RSV vaccine (ChAd155-RSV vaccine) have been evaluated in healthy adults aged 18 to 45 years (study 201974 [RSV PED-001; NCT02491463]) There were no significant safety concerns identified up to Day 60 in study RSV PED-001. Overall, the ChAd155-RSV vaccine high dose (Hd) (5 x 10^{10} vp) seems to be more reactogenic (local and general) than the ChAd155-RSV vaccine low dose (Ld) (5 x 10^{9} vp), however, the reactogenicity profile was less than that observed in the Bexsero group. No safety signal from the assessed hematology parameters (hemoglobin, platelet count, prothrombin time and APTT) was observed in subjects receiving the ChAd155-RSV vaccine. No significant reductions in platelet count or clinically significant changes in coagulation parameters were observed up to 30 days post Dose 2. An approximately 2.4-fold increase in RSV-A neutralizing antibody titers (geometric mean titer [GMT] from baseline) was observed in both RSV-Ld and RSV-Hd after Dose 1. No booster effect was evident after Dose 2. An anti-F IgA and IgG antibody secreting B-cells response and an RSV F, N and M2-1 specific IFN-γ secreting T-cells response in RSV-Hd group after the first dose were observed with ELISpot. There was no booster response after the second vaccination. There was no specific vaccine-induced CD4 T-cellular response observed with intracellular staining (ICS). For CD8 T-cells only a weak CD8 IFN-γ response to N was shown with ICS for some subjects.

Please refer to the current IB for information regarding the pre-clinical ***and clinical*** studies of GlaxoSmithKline (GSK) Biologicals' investigational ChAd155-RSV vaccine.

In Section 1.2.2.1. Study population, the following changes have been made:

The immunogenicity, safety and reactogenicity of the pediatric candidate RSV vaccine (ChAd155-RSV vaccine) ~~are currently being~~ ***has been*** evaluated in healthy adults aged 18 to 45 years (study 201974 [RSV PED-001; NCT02491463]).

In Section 1.2.3. Rationale for regimen, dose and route of administration, the following changes have been made:

Please refer to the current IB for information regarding the pre-clinical***, clinical*** and toxicology studies of GSK Biologicals' investigational ChAd155-RSV vaccine.

In Section 1.2.4.1. Staggered design with 3 steps, the following changes have been made:

~~Within each step, a~~***A***n internal Safety Review Committee (iSRC) will review ***all accumulating*** ~~accumulated~~ safety data three weeks after the start of vaccination and then about every three weeks until the IDMC ~~reviews~~ ***has reviewed all safety data up to 30 days after administration of Dose 2 in Step 3 (i.e. Day 60)*** ~~the data~~. Ad-hoc iSRC meetings may be held should safety concerns warrant a safety review. Refer to Section 9.10.2 for more information about iSRC.

The IDMC will review accumulating reactogenicity and safety data monthly throughout the period of vaccination (up to 30 days after administration of Dose 2 in Step 3, i.e. Day 60). The IDMC will also review incidence of all serious adverse events (SAEs) and incidence of RSV-respiratory tract infections (RTI), RSV-LRTI, RSV-severe LRTI and RSV-RTI leading to hospitalization monthly after the start of vaccination and from beginning of the first RSV season to the end of the study.

Both iSRC and IDMC will receive critical safety data (i.e. SAEs and adverse events of specific interest) within 48 hours upon GSK becoming aware of it.

The iSRC and IDMC will determine whether any of the predefined study holding rules are met. If this is the case, vaccination in the study will be immediately put on hold. At any time, the IDMC may recommend to the sponsor to suspend the enrolment to the study and/or vaccination based on the safety data regularly arising in this study.

~~Moving to a next step will be conditional to the favorable outcome of the evaluation of safety data obtained up to 7 days after administration of the second vaccine dose in the previous step (for all subjects), performed by an IDMC.~~ ***Dose escalation will proceed after administration of the vaccine to all 32 subjects in the previous Step, and in the absence of a safety concern detected by the IDMC in the regular monitoring of accumulating safety data. It will be ensured that, as a minimum the IDMC will have reviewed safety data of at least 16 subjects for two doses and given their formal approval prior to enrolling subjects to the next dose level.***

In Section 1.2.4.2. Rationale for monitoring of hematology parameters, the following changes have been made:

To stay within blood sampling limits that are consistent with physiological "minimal risk" in this young and vulnerable population, but also to minimize any development of needle aversion, ~~no blood draw will be performed for immunological assays and for biochemistry tests at Day 1 and none for hematology and biochemistry tests at Day 3~~ ***blood draws for the determination of hematological parameters are scheduled at Day 1 and Day 31 (corresponding to the first day after administration of the vaccine). Additional specimens will be drawn if any ≥ Grade 1 for platelet decrease is detected at Day 1 or Day 31 to investigate clinical symptoms suggestive of low platelets.*** Blood samples collected within the screening window will serve as baseline for the immunological analysis and for the hematology and biochemistry check-up before vaccination. Consequently, the maximum blood draw volumes collected per protocol have been restricted to 9.0 mL within 24 hours and ~~32.4~~ ***26.2*** mL over eight weeks (refer to Table 5 and Table 7). ***For investigational sites without a laboratory in proximity capable to perform whole blood stimulation [WBS] necessary for the CMI assay the volumes will be less.***

In light of these data, a clinical examination with special attention given to the detection of spontaneous bleeding or easy bruising and evidence of bruising or petechiae will be performed ***by the investigator/study staff*** on Day 1, Day 3, ~~and~~ ***Day 7, Day 31, Day 33 and Day 37*** ~~after each vaccination~~. ***The visits on Day 3 and Day 33 may be replaced by a phone call by the investigator/study staff to inquire of the infants' general health and to solicit reports of a rash, spontaneous bleeding or easy bruising.*** On Day 1 after each

vaccination, special attention will also be given to the detection of petechiae or evidence of petechiae induced by the tourniquet. ***This special attention to detection of petechiae or evidence of petechiae induced by the tourniquet will also be given on Day 7 and Day 37 in case hematology testing will be performed on that day***. Subjects' parent(s)/ legally acceptable representative***(s)*** (LAR[s]) will be instructed to contact the investigator/study staff if their child presents symptoms of spontaneous bleeding or easy bruising ***or if their child develops a rash in the month*** following vaccination***, in order to detect any thrombocytopenic petechiae or purpura. The investigator will, based on his/her medical judgement, appropriately investigate infants with clinical suspicion of low platelets.***

Note that a broad safety hematology and biochemistry evaluation was performed at each study visit in adults during study RSV PED-001 (201974; NCT02491463). In this study, blood samples for hematology/biochemistry were taken from all subjects at all study visits, i.e. at Screening, on the day of vaccination (Day 0 and Day 30), 1 day post vaccination (Day 1 and Day 31), 3 days post vaccination (Day 3 and 33), 7 days post vaccination (Day 7 and 37), 30 days post Dose 2 (Day 60), on Day 180 and on Day 360. The safety profile of the ChAd155-RSV vaccine was found to be satisfactory by an IDMC. No safety signal from the assessed hematology parameters (hemoglobin, platelet count, prothrombin time and APTT) was observed in subjects receiving the ChAd155-RSV vaccine. No significant reductions in platelet count or clinically significant changes in coagulation parameters were observed up to 30 days post Dose 2 (refer to Section 1.1.6 and current IB).

In Section 1.3.1. Risk Assessment, the following changes have been made:

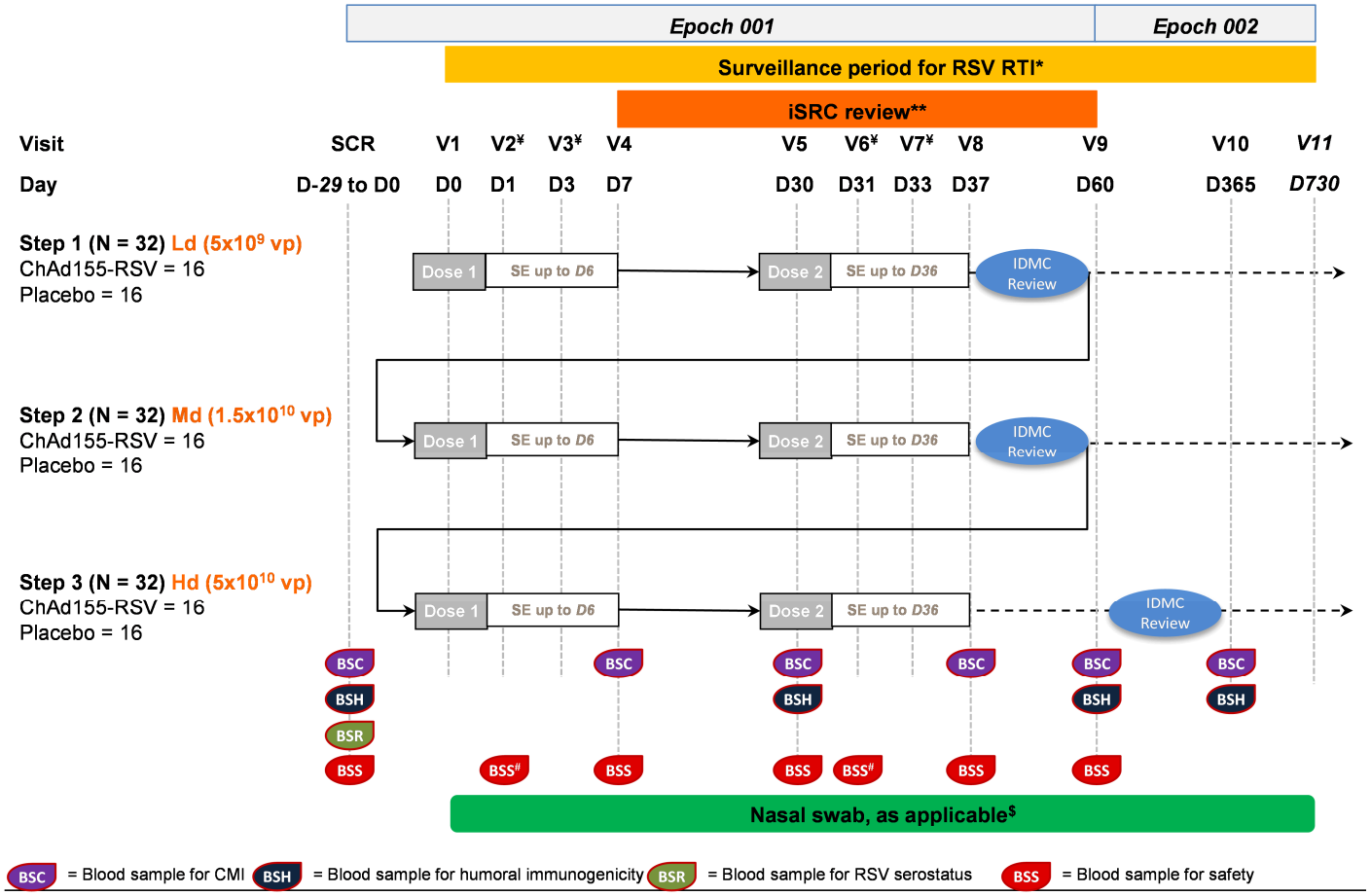
No significant safety concerns were identified up to 30 days post Dose 2 in study RSV PED-001 with the ChAd155-RSV vaccine in healthy adults aged 18 to 45 years (refer to Section 1.1.6 and to the current IB).

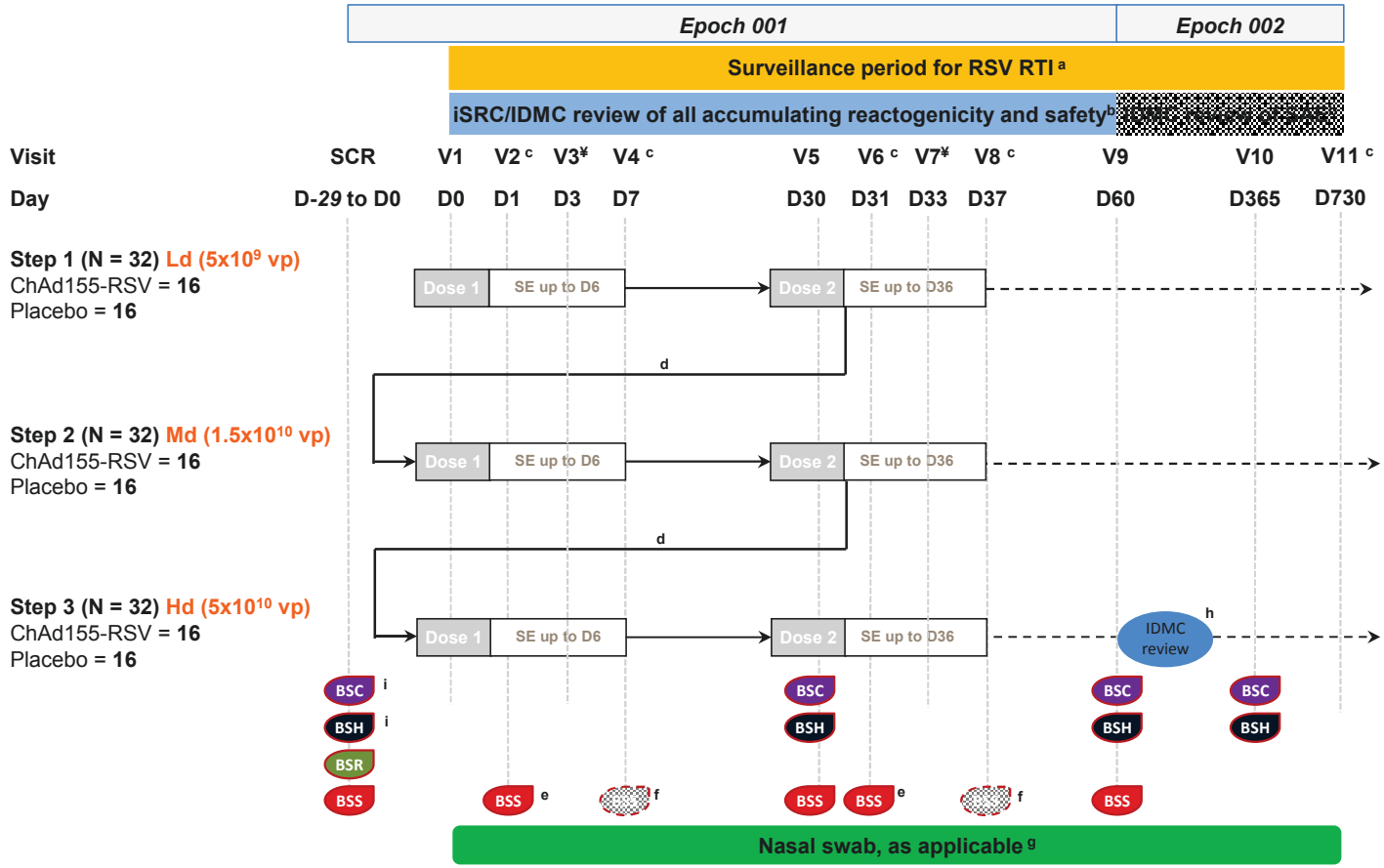
In Section 2.3. Tertiary objectives, the following changes have been made:

- To further evaluate the CMI ***profile*** induced by two IM doses of three dose levels of the RSV investigational vaccine when administered according to a 0, 1-month schedule, up to Day 365 in RSV-seropositive infants aged 12 to ~~17~~***23*** months.

In Section 3. Study Design Overview, the following changes have been made:

Figure 1 Study design





BSC = Blood sample for CMI BSH = Blood sample for humoral immunogenicity BSR = Blood sample for RSV serostatus BSS = Blood sample for safety

SE: ~~safety evaluation~~ ***solicited events***;

¥ On Day 3 and Day 33, a visit may take place at the investigators clinical facility or the investigator/clinical staff may call the subjects' parent(s)/LAR(s), as appropriate to the circumstances in the judgment of the investigator.

a Surveillance for RSV-RTI comprises monthly nasal swab collected to detect asymptomatic RSV infections during RSV season and active and passive surveillance contacts for RSV symptomatic RTI (see Section 9.2). ***Data about RSV–RTI incidence will be reviewed monthly by an IDMC.***

b Within each step, an iSRC will review ~~accumulated~~ ***all accumulating*** safety data three weeks after the start of vaccination and then about every three weeks (until the IDMC ~~reviews the data.)~~ ***has reviewed all safety data up to 30 days after administration of Dose 2 in Step 3, i.e. Day 60)***. ***The IDMC will review all accumulating reactogenicity and safety data monthly throughout the period of vaccination (up to 30 days after administration of Dose 2 in Step 3, i.e. Day 60) and accumulating SAEs until Day 730. Refer to Sections 9.10.2 and. 9.10.3.***

c Visit 2 (Day 1), ~~Visit 3 (Day 3)~~***, Visit 4 (Day 7),*** Visit 6 (Day 31), ~~Visit 7 (Day 33),~~ ***Visit 8 (Day 37)*** and Visit 11 (Day 730) may take place in the subject's home or at the investigators clinical facility as appropriate to the circumstances in the judgment of the investigator.

d Dose escalation will proceed after administration of the vaccine to all 32 subjects in the previous Step, and in the absence of a safety concern detected by the IDMC in the regular monitoring of accumulating safety data. It will be ensured that, as a minimum the IDMC will have reviewed safety data of at least 16 subjects for two doses and given their formal approval prior to enrolling subjects to the next dose level.

e Only for hematology.

f Day 7 hematology testing will be performed if any ≥ Grade 1 for platelet decrease is detected on Day 1 after vaccination; to ensure its resolution. Similarly, hematology testing on Day 37 will be performed if any ≥ Grade 1 for platelet decrease is detected on Day 31.

g Refer to Section 6.6.11.3.

h The IDMC will perform a review when all safety data up to 30 days after administration of Dose 2 in Step 3 (i.e. Day 60) are available.

i Blood samples for CMI and for humoral immunogenicity may be postponed until after the RSV serostatus is known and eligibility is confirmed up to or on Day 0 (but before vaccination).

- End of Study: Last testing results released of samples collected at Visit 11 (Day 730)***.****

  **** Up to Visit 11 (Day 730), there will be monthly nasal swab to detect asymptomatic RSV infections during the RSV season and if following active or a passive surveillance contacts, a subject presents symptoms of RTI, a nasal swab will be collected.***

- Sampling schedule:

  – Blood samples for **biochemistry** will be taken from all infants at Screening (up to 29 days before first vaccination to Day 0) and on ~~Day 7,~~ Day 30, ~~Day 37~~, and Day 60 (refer to Table 11 for the list of parameters to be tested). Blood samples for **hematology** will be taken from all infants at Screening (up to 29 days before first vaccination to Day 0) and on Day 1, ~~Day 7,~~ Day 30, Day 31, ~~Day 37,~~ and Day 60 (refer to Table 11 for the list of parameters to be tested). ***Day 7 hematology testing will be performed if any ≥ Grade 1 for platelet decrease is detected on Day 1 after vaccination, to ensure its resolution. Similarly, hematology testing on Day 37 will be performed if any ≥ Grade 1 for platelet decrease is detected on Day 31.*** A clinical ~~history and~~ examination with special attention given to detection of spontaneous bleeding or easy bruising and evidence of bruising or petechiae will be performed ***by the investigator/study staff*** on Day 1, Day 3 and Day 7 after each vaccination (i.e. Day 1, Day 3, Day 7, Day 31, Day 33, and Day 37). ***The visits on Day 3 and Day 33 may be replaced by a phone call by the investigator/study staff to inquire of the infants' general health and to solicit reports of a rash, spontaneous bleeding or easy bruising.*** On Day 1, ~~Day 7,~~ ***and*** Day 31, ~~and Day 37,~~ special attention will also be given to the detection of petechiae or evidence of petechiae induced by the tourniquet. ***This special attention to detection of petechiae or evidence of petechiae induced by the tourniquet will also be given on Day 7 and/or Day 37 in case hematology testing will be performed on that day***. Further testing may be required to investigate a finding or guide subject management based on the investigators clinical judgment. Subjects' parent(s)/LAR(s) will be instructed to contact the investigator/study staff if their child presents symptoms of spontaneous bleeding or easy bruising ***or if their child develops a rash in the month*** following vaccination***, in order to detect any thrombocytopenic petechiae or purpura. The investigator will, based on his/her medical judgement, appropriately investigate infants with clinical suspicion of low platelets.***

  – Blood samples for **CMI** ***are limited to those in investigational sites with a laboratory in proximity capable to perform WBS necessary for the CMI assay. In those sites, samples*** will be taken from all subjects at Screening ~~***(or alternatively during an additional visit before Day 0)***~~ and on ~~Day 7,~~ Day 30, ~~Day 37,~~ Day 60, and Day 365**.**

    Note: Blood samples for CMI taken on Day 7 and Day 37 for subjects enrolled under protocol amendment 1 will still be assessed for CMI.

- Blood samples for **humoral immunogenicity** will be taken from all subjects at Screening ~~***(or alternatively during an additional visit before Day 0)***~~ and on Day 30, Day 60, and Day 365.

- ***Study visits: Visit 1 (Day 0), Visit 5 (Day 30), Visit 9 (Day 60) and Visit 10 (Day 365) must be performed at the investigators clinical facility. Visit 2 (Day 1), Visit 4 (Day 7), Visit 6 (Day 31), Visit 8 (Day 37) and Visit 11 (Day 730) (no blood sampling for immune response and no vaccine administration) may take place in the subject's home or at the investigators clinical facility as appropriate to the circumstances in the judgment of the investigator. Visit 3 (Day 3) and Visit 7 (Day 33) may take place at the investigators clinical facility or the investigator/clinical staff may call the subjects' parent(s)/LAR(s), as appropriate to the circumstances in the judgment of the investigator.***

In Section 4.1 RTI case definitions, the following changes have been made:

During the analysis of the study, all cases identified during the surveillance of RSV-RTI will be definitively classified as either RTI, LRTI, ~~and~~ severe LRTI ***or very severe LRTI*** according to the ***standardized*** case definitions ~~described in~~ ***(see*** Table 4***) based on the available World Health Organization (WHO) case definitions***. ~~The analysis may also be done using the available World Health Organization (WHO) case definitions for RTI, LRTI and severe LRTI.~~

Table 4 Case definitions for data analysis

RSV-RTI	**RSV-LRTI***	**severe RSV-LRTI***
RSV A or B positive by quantitative PCR AND Runny nose, OR Blocked nose, OR Cough	RSV A or B positive by quantitative PCR AND Infant with RTI AND SpO_2 < 95% (measured in room air by pulse oximetry), OR RR increase: > 40/minutes 12 - 30 months of age	RSV A or B positive by quantitative PCR AND Infant with LRTI AND SpO_2 < 92% (measured in room air by pulse oximetry), OR Difficulty breathing leading to: Irritability/agitation, OR Lethargy/sleepiness, OR Lower chest wall indrawing, OR Reduced/no vocalization, OR Apnea > 20 seconds, OR Cyanosis, OR Stop feeding well/dehydration
All-cause RTI	**All-cause LRTI**	**All-cause severe LRTI**
Runny nose, OR Blocked nose, OR Cough	Infant with RTI AND SpO_2 < 95% (measured in room air by pulse oximetry), OR RR increase: > 40/minutes 12 - 30 months of age	Infant with LRTI AND SpO_2 < 92% (measured in room air by pulse oximetry), OR Difficulty breathing leading to: Irritability/agitation, OR Lethargy/sleepiness, OR Lower chest wall indrawing, OR Reduced/no vocalization, OR Apnea > 20 seconds, OR Cyanosis, OR Stop feeding well/dehydration

PCR: polymerase chain reaction; **RR**: respiratory rate; **SpO_2**: blood oxygen saturation; **RTI**: respiratory tract infections; **LRTI**: lower respiratory tract infections

* Any infants meeting the RSV-LRTI case definition will be included in the RSV-RTI case definition and any infants meeting the severe RSV-LRTI case definition will be included in the RSV-LRTI and RSV-RTI case definitions.

RSV-RTI	***Runny nose OR blocked nose OR cough*** ***AND*** ***Confirmed RSV infection [4]***
RSV-LRTI	***History of cough OR difficulty breathing [1]*** ***AND*** ***SpO_2 < 95% [2], OR RR increase [3]*** ***AND*** ***Confirmed RSV infection [4]***
RSV-severe LRTI	***Meeting the case definition of RSV-LRTI*** ***AND*** ***SpO_2 < 93% [2], OR lower chest wall in-drawing***
RSV-very severe LRTI	***Meeting the case definition of RSV-LRTI*** ***AND*** ***SpO_2 < 90% [2], OR inability to feed, OR failure to respond / unconscious***
RSV hospitalization	***Confirmed RSV infection [5]*** ***AND*** ***Hospitalized for acute medical condition [6]***
All-cause LRTI	***History of cough OR difficulty breathing [1]*** ***AND*** ***SpO_2 < 95% [2], OR RR increase [3]***

Definitions based on [Modjarrad, 2016]
LRTI = lower respiratory tract infections; RR = respiratory rate; RTI = respiratory tract infections; SpO_2 = blood oxygen saturation.
[1] Based on history reported by parents/LARs and includes difficulty breathing (e.g. showing signs of wheezing or stridor, tachypnoea, flaring [of nostrils], chest in-drawing, apnoea) associated with nasal obstruction.
[2] For blood oxygen saturation (SpO_2), the lowest value monitored will be used.
[3] RR increase defined as ≥ 40/minute (12 months of age or above).
[4] RSV infection confirmed on nasal swab positive for RSV A or B by quantitative Reverse Transcription Polymerase Chain Reaction (qRT-PCR).
[5] RSV sampling and testing is based on medical judgment of medical practitioner or driven by algorithm.
[6] Hospitalization is defined as a medical decision that the infant requires admission for observation or treatment.

In Section 5.2. Inclusion criteria, the following changes have been made:

- ~~Born full-term (i.e. after a gestation period of 37 to less than 42 completed weeks) with a minimum birth weight of 2.5 kg.~~

In Section 6.5. Outline of study procedures, the following changes have been made:

Table 5 List of study procedures

Epoch	Epoch 001										Epoch 002					
Age	**12-~~17~~*23* mth**										**24-~~29~~*35* mth**	**36-~~41~~ *47* mth**				
Type of contact	**Screening**	**Visit 1**	**Visit 2#**	**Visit 3##**	**Visit 4#**	**Visit 5**	**Visit 6#**	**Visit 7##**	**Visit 8#**	**Visit 9**	**Visit 10**	**Visit 11#**	**Unscheduled visit for safety***	**Contact for active/passive surveillance****	**Surveillance for asymptomatic RSV-RTI*****	**Assessment of potential RSV-RTI********
Timepoints	**D-29 to D0**	**D0**	**D1**	**D3**	**D7**	**D30**	**D31**	**D33**	**D37**	**D60**	**D365**	**D730**		**Monthly or Weekly**	**Monthly**	
Informed consent	●															
~~Informed consent Addendum[$]~~	~~●~~															
Check inclusion/exclusion criteria	●	O														
Collect demographic data	●[a]															
Medical history	●															
Physical examination[b]	●	O	O	~~O~~	O	O	O	~~O~~	O	O	O	O	O		O	●
Growth monitoring[c]	●					●				●	●	●				
Check contraindications and warnings and precautions		O				O										
Pre-vaccination body temperature		●				●										
Randomization		O														
Vaccine administration		●				●										
Recording of administered treatment number		●				●										
60 minutes post-vaccination observation[d]		●				●										
Blood sampling for RSV serostatus (1.0 mL)	●															
Blood sampling for assessment of mechanisms of illness (potential ERD; 2.5 mL)																●[l]

Epoch	Epoch 001										Epoch 002					
Age	12-~~17~~*23* mth										~~24-29~~*35* mth	36-~~41~~ *47* mth				
Type of contact	Screening	Visit 1	Visit 2[#]	Visit 3[##]	Visit 4[#]	Visit 5	Visit 6[#]	Visit 7[##]	Visit 8[#]	Visit 9	Visit 10	Visit 11[#]	Unscheduled visit for safety*	Contact for active/passive surveillance**	Surveillance for asymptomatic RSV-RTI***	Assessment of potential RSV-RTI****
Timepoints	D-*29* to D0	D0	D1	D3	D7	D30	D31	D33	D37	D60	D365	D730		Monthly or Weekly	Monthly	
Blood sampling for hematology (1.2 mL)	●[e]		●[f]		●[f,*p*]	●[f,g]	●[f]		●[f,*p*]	●[f]			●[f]			
Blood sampling for biochemistry (1.1 mL)	●[e]				●[f,]	●[f,g]			●[f,]	●[f]			●[f]			
Blood sampling for CMI response (2.0 mL)[o]	●[*r*]				●	●[g]			●	●	●					
Blood sampling for humoral response (2.5 mL)	●[*r*]					●[g]				●	●					
Detection of spontaneous bleeding or easy bruising and evidence of bruising or petechiae[q]			●	●	●		●	●	●							
Examination of limb for petechiae induced by the tourniquet			●		●		●		●							
Surveillance for RSV-RTI, difficulty in breathing and wheezing		O	O	O	O	O	O	O	O	O	O	O		●	●	
Documentation of symptoms and signs of RTI[j]																●
Nasal swab for central testings															●[m]	●[h]
Specimen for local testings																●[n]
Distribution of RTI episode cards[k]		O														
Collection of completed RTI episode cards[k]			O	~~O~~	O	O	O	~~O~~	O	O	O	O	O		O	O
Transcription of completed RTI episode cards			●	●	●	●	●	●	●	●	●	●	O		●	●
Record any concomitant medications/vaccinations		●	●	●	●	●	●	●	●	●	●	●	●		●	●
Distribution of the subject card		O														

Epoch	Epoch 001										Epoch 002					
Age	**12-~~17~~*23* mth**										**24-~~29~~*35* mth**	**36-~~41~~ *47* mth**				
Type of contact	**Screening**	**Visit 1**	**Visit 2#**	**Visit 3##**	**Visit 4#**	**Visit 5**	**Visit 6#**	**Visit 7##**	**Visit 8#**	**Visit 9**	**Visit 10**	**Visit 11#**	**Unscheduled visit for safety***	**Contact for active/passive surveillance****	**Surveillance for asymptomatic RSV-RTI*****	**Assessment of potential RSV-RTI******
Timepoints	**D-*29* to D0**	**D0**	**D1**	**D3**	**D7**	**D30**	**D31**	**D33**	**D37**	**D60**	**D365**	**D730**		**Monthly or Weekly**	**Monthly**	
Distribution of diary card		O				O										
Return of diary card					O	O			O	O						
Diary card transcription by investigator					●	●			●	●						
Recording of solicited AEs (Day 0-6)		●	●	~~●~~	●	●	●	~~●~~	●				●[i]			
Recording of unsolicited AEs (Day 0-29)		●	●	●	●	●	●	●	●	●			●[i]			
Recording of AE leading to study withdrawal		●	●	●	●	●	●	●	●	●	●	●	●	●	●	●
Recording of AE of specific interest (RSV-LRTI)		●	●	●	●	●	●	●	●	●	●	●	●	●	●	●
Recording of AE of specific interest (spontaneous or excessive bleeding)		●	●	●	●	●	●	●	●	●			●	●	●	●
Recording of SAEs		●	●	●	●	●	●	●	●	●	●	●	●	●	●	●
Recording of SAEs related to study participation, or to a concurrent GSK medication/vaccine	●	●	●	●	●	●	●	●	●	●	●	●	●	●	●	●
Screening conclusion	●															
Study conclusion												●				
~~Study conclusion for GSK data management~~ ***Signing of investigator signature form by investigator after Screening and before each analysis***	●									●	●	●				

* This visit is applicable for infants with hematological/biochemical values out of normal range ***or for further evaluation of clinical suspicion of low platelet count.***

** Active contacts for surveillance of RSV-RTI will take place weekly during the RSV season and every month outside the RSV season (refer to Section 9.2). Passive phone contacts from the ~~carers~~ ***subjects' parent(s)/LAR(s)*** to the investigator will take place when symptoms occur.

Visit 2 (Day 1), ~~Visit 3 (Day 3)~~***,Visit 4 (Day 7),*** Visit 6 (Day 31), ~~Visit 7 (Day 33),~~ ***Visit 8 (Day 37)*** and Visit 11 (Day 730) may take place in the subject's home or at the investigators clinical facility as appropriate to the circumstances in the judgment of the investigator.

[##] ***On Day 3 and Day 33, a visit may take place at the investigators clinical facility or the investigator /clinical staff may call the subjects' parent(s)/LAR(s), as appropriate to the circumstances in the judgment of the investigator.***

~~$ This is only applicable for subject's parent(s)/LAR(s) who have previously signed the Version 1~~***2*** ~~of the model ICF~~

[b] ***At Screening, p***~~P~~perform a complete physical examination, including assessment of vital signs (body temperature, heart rate [HR], and RR). ***At subsequent study visits, perform a physical examination only if the subject's parent(s)/LAR(s) indicate(s) during questioning that there might be some underlying pathology(ies) or if deemed necessary.***

[e] At Screening, for infants with hematological/biochemical values out of normal range which are expected to be temporary, a re-screening visit may be scheduled during which blood sample collection for hematology/biochemistry will be repeated (maximum one re-screening visit per infant is allowed; ***blood for CMI response and humoral response will not be re-sampled at the re-screening visit*****), if already done before**.

[f] If any Grade 1 abnormality with potential clinical relevance (according to investigators judgment) or any ≥ Grade 2 abnormality is detected***, or for further evaluation of clinical suspicion of low platelet count,*** refer to Section 6.6.11.2 and Figure 2 for re-test.

[h] If during passive or active surveillance contact, the investigator/study staff assesses that an infant presents a potential RSV-RTI (refer to Table 4 for definition), the potential RSV infection will be assessed by quantitative ***RT-***PCR.

[m] Nasal swab collected at the monthly surveillance for asymptomatic RSV-RTI will be tested by quantitative ***RT-***PCR.

[o] ***In investigational sites with a laboratory in proximity capable to perform WBS necessary for the CMI assay.***

[p] ***Day 7 hematology testing will be performed if any ≥ Grade 1 for platelet decrease is detected on Day 1 after vaccination. Similarly, hematology testing on Day 37 will be performed if any ≥ Grade 1 for platelet decrease is detected on Day 31***

[q] ***A clinical examination with special attention given to detection of spontaneous bleeding or easy bruising and evidence of bruising or petechiae will be performed by the investigator/study staff on Day 1, Day 3 and Day 7 after each vaccination (i.e. Day 1, Day 3, Day 7, Day 31, Day 33 and Day 37). The visits on Day 3 and Day 33 may be replaced by a phone call by the investigator/study staff to inquire of the infants' general health and to solicit reports of a rash, spontaneous bleeding or easy bruising. Subjects' parent(s)/LAR(s) will be instructed to contact the investigator/study staff if their child presents symptoms of spontaneous bleeding or easy bruising or if their child develops a rash in the month following vaccination, in order to detect any thrombocytopenic petechiae or purpura.***

[r] ***Blood samples for CMI and for humoral immunogenicity may be postponed until after the RSV serostatus is known and eligibility is confirmed up to or on Day 0 (but before vaccination).***

Table 6 Intervals between study visits

Interval	Optimal length of interval	Allowed interval
Visit 1 (Day 0) → Visit 4 (Day 7)	7 days	7 - ~~9~~***10*** days[2]
Visit 5 (Day 30) → Visit 8 (Day 37)	7 days	7 - ~~9~~***10*** days[2]

[1] Visit 1 should take place no longer than 30 days after the Screening visit. When applicable, a re-screening visit may be scheduled at any time (but only once to assess eligibility; blood for CMI response and humoral response will not be re-sampled ***if already done before***). All screening procedures need to be performed within 30 days of Visit 1.
[2] Immunogenicity data from blood samples collected outside this interval will not be eligible for inclusion in the according-to-protocol (ATP) cohort for analysis of immunogenicity.

In Section 6.6.5. Physical examination, the following changes have been made:

At screening, ~~P~~***p***erform a physical examination of the subject, including assessment of vital signs preferably when the infant is calm. Vital signs are body temperature, heart rate (HR) and RR. Collected information needs to be recorded in the eCRF.

In Section 6.6.11.1. Blood sampling for RSV serostatus, humoral immunity, and cell-mediated immunity, the following changes have been made:

- A volume of 2.0 mL of whole blood should be drawn from all infants for analysis of CMI response at each pre-defined timepoint***, in investigational sites with a laboratory in proximity capable to perform WBS necessary for the CMI assay.*** Refer to the SPM for more details on sample storage conditions.
- ***Blood sampling for CMI and humoral response will not be repeated in case of insufficient volume, at any timepoint.***

In Section 6.6.11.2. Blood sampling for hematology and biochemistry, the following changes have been made:

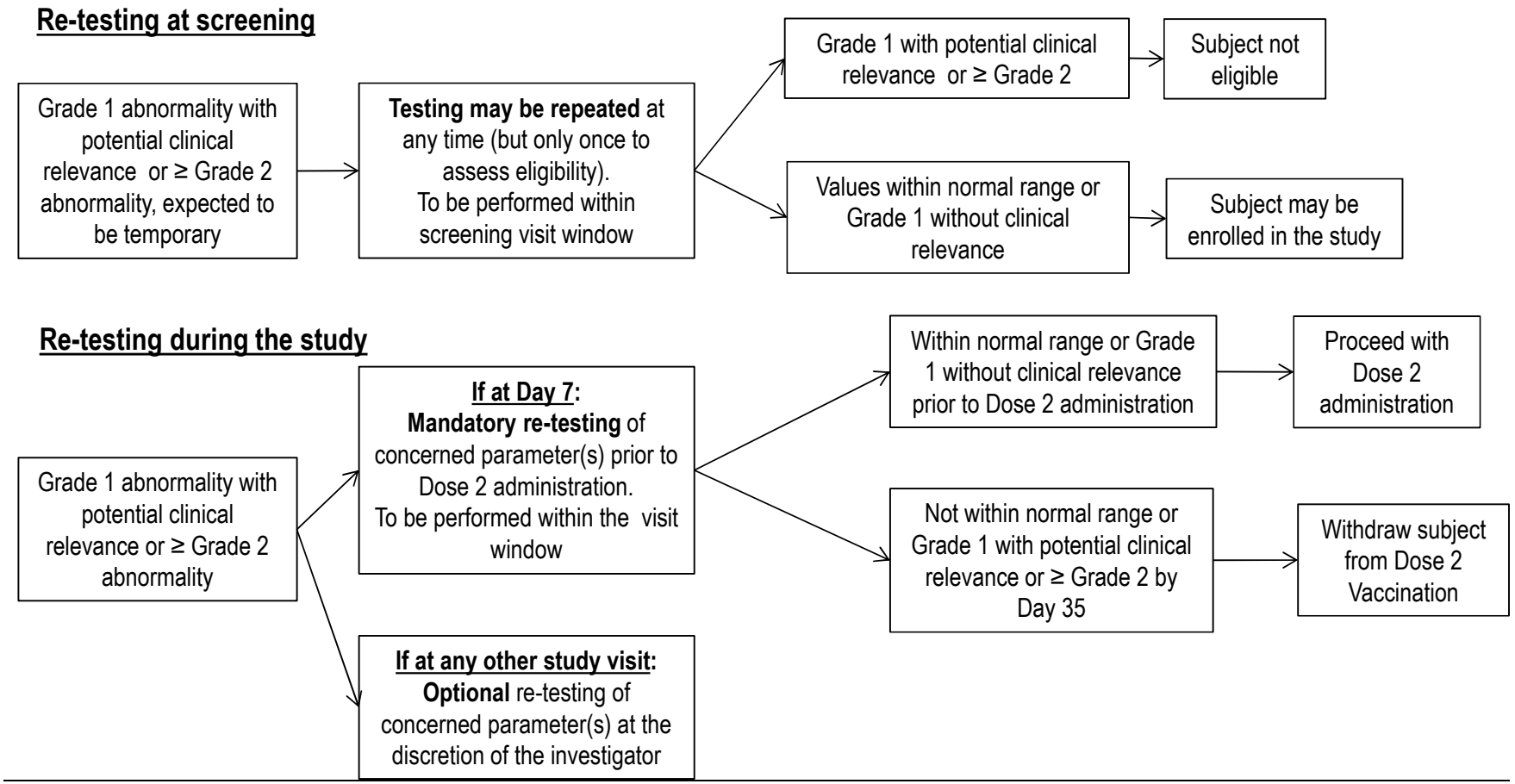
Day 7 hematology testing will be performed if any ≥ Grade 1 for platelet decrease is detected on Day 1 after vaccination. Similarly, hematology testing on Day 37 will be performed if any ≥ Grade 1 for platelet decrease is detected on Day 31 (refer to Figure 2 and Appendix C).

If any Grade 1 abnormality with potential clinical relevance (according to investigators judgment, refer to the glossary of terms) or any ≥ Grade 2 abnormality is detected (refer to Figure 2 and Appendix C):

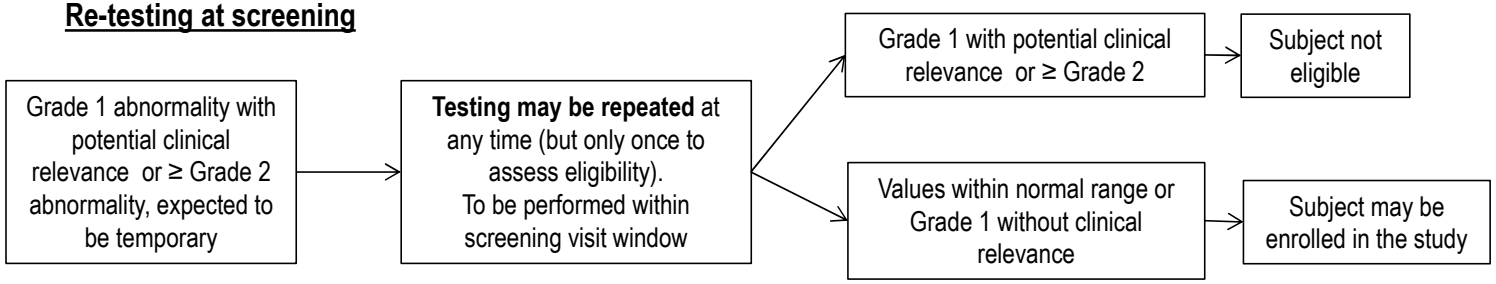
- **at Day 7**, the relevant hematology/biochemistry parameter(s) **must** be re-tested before administration of vaccine Dose 2, either during an unscheduled visit or at the Dose 2 vaccination visit itself (provided results are obtained before vaccination). Only if the concerned parameter(s) is/are within the acceptable range (i.e. within normal range or Grade 1 without clinical relevance), vaccine Dose 2 can be administered. If the concerned parameter(s) is/are not within the acceptable range ~~by Day 35 (which is the maximum~~ ***within the*** allowed interval for the second dosing visit, Visit 5), the infant will not receive Dose 2 but should still continue the study for safety follow-up (up to Day 730).
- **at Days ~~1,~~ 30, ~~31,~~ 37 and 60** the relevant hematology/biochemistry parameter(s) **may** be re-tested as per the opinion of the investigator.

Note: For safety reasons and specifically for further investigation of an AE of specific interest (suspected bleeding or low platelet count), blood can be re-sampled for hematology and biochemistry assessment.

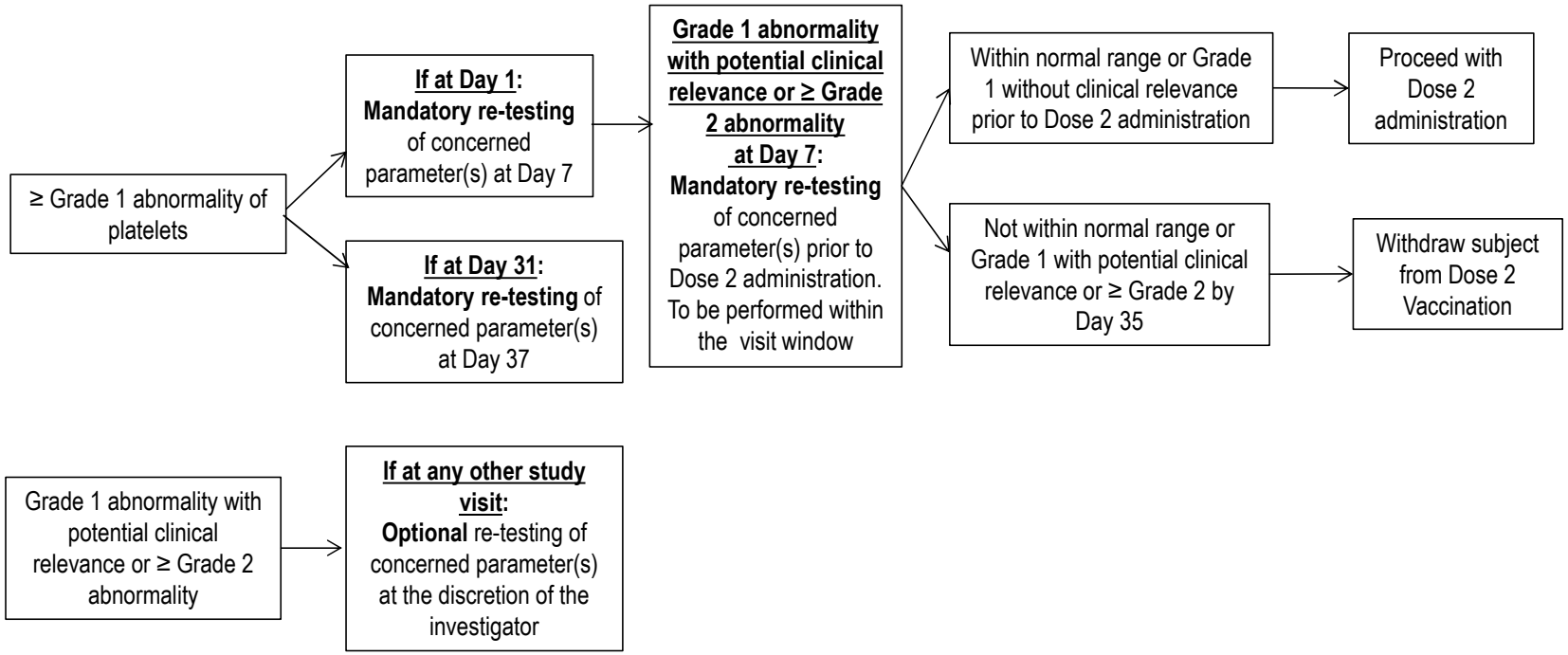
Figure 2 Schematic overview of required additional hematology and biochemistry testing during the study



Re-testing at screening



Re-testing during the study



Note: For safety reasons and specifically for further investigation of an AE of specific interest (suspected bleeding or low platelet count), blood can be re-sampled for hematology and biochemistry assessment.

In Section 6.6.11.3. Nasal swab and other specimen for local assay, the following changes have been made:

Nasal swab and other specimen for local assay collected during assessment visit

If during passive or active surveillance contact, the investigator/study staff assesses that an infant presents a potential RSV-RTI (refer to 9.2), a nasal swab and another specimen for local assay will be collected during an assessment visit:

- The nasal swab will allow ***assessing*** the potential RSV infection by quantitative ***RT-***PCR at sponsor laboratory.
- The specimen for local assay will allow ***assessing*** the potential RSV infection and RVP at a local routine laboratory, where available. The specimen type will depend on the assay run locally.

Nasal swab collected to detect asymptomatic RSV infections

During RSV season, there will be monthly nasal swabs to detect asymptomatic RSV infections. These nasal swab***s*** will allow ***assessing*** the potential RSV infection by quantitative ***RT-***PCR at sponsor laboratory.

~~Cells and secretions from the nasal septum will be collected using sterile swabs.~~ Refer to the SPM for more details about nasal swab.

In Section 6.6.12. Detection of spontaneous bleeding or easy bruising and evidence of bruising or petechiae, the following changes have been made:

Subjects' parent(s)/LAR(s) will be instructed to contact the investigator/study staff if their child presents symptoms of spontaneous bleeding or easy bruising ***or if their child develops a rash in the month*** following vaccination***, in order to detect any thrombocytopenic petechiae or purpura. Refer to Sections 9.1.5 and 9.4.3 for reporting of adverse events of specific interest (spontaneous or excessive bleeding).***

At Visit 2 (Day 1), Visit 3 (Day 3)*, Visit 4 (Day 7), Visit 6 (Day 31), Visit 7 (Day 33)*, and Visit 8 (Day 37), the investigator will perform a clinical examination with special attention given to detection of spontaneous bleeding or easy bruising and evidence of bruising or petechiae. Episodes of spontaneous bleeding or easy bruising and evidence of bruising or petechiae will be recorded in the eCRF.

**** The visits on Day 3 and Day 33 may be replaced by a phone call by the investigator/study staff to inquire of the infants' general health and to solicit reports of a rash, spontaneous bleeding or easy bruising.***

In Section 6.6.13. Detection of petechiae or evidence of petechiae induced by the tourniquet, the following changes have been made:

At Visit 2 (Day 1), ~~Visit 4 (Day 7),~~ Visit 6 (Day 31), ~~and Visit 8 (Day 37),~~ the investigator will perform an examination of the limb where the tourniquet for blood sample has been placed in order to detect petechiae or evidence of petechiae induced by

the tourniquet. ***This special attention to detection of petechiae or evidence of petechiae induced by the tourniquet will also be given at Visit 4 (Day 7) and/or Visit 8 (Day 37) in case hematology testing will be performed on that day***. Episodes of petechiae will be recorded in the eCRF.

In Section 6.6.14. Surveillance for RSV-RTI and wheezing, the following changes have been made:

At the first vaccination visit (Visit 1 [Day 0]), RTI episode card***s*** will be provided to the subject's parent(s)/LAR(s). ***At subsequent study visits, new RTI episode card(s) will be distributed as needed.***

In Section 6.6.18. Signing of investigator signature form by investigator, the following changes have been made:

At the end of the screening visit, Visit 9 (Day 60), Visit 10 (Day 365) and Visit 11 (Day 730), the investigator will sign the investigator signature form in the eCRF.

In Section 6.7.2. Biological samples, the following changes have been made:

Table 7 Biological samples

Sample	Timepoint	Type of sample †	Subject	Number of subjects	Quantity
Blood	Screening	Blood sample for RSV serostatus	All infants	≥ 96	1.0 mL
		Blood sample for hematology	All infants	≥ 96	1.2 mL
		Blood sample for biochemistry	All infants	≥ 96	1.1 mL
		Blood sample for cell-mediated immunogenicity response #	All infants††	≥ ***or*** ≤96 ††,¥	2.0 mL
		Blood sample for humoral response #	All infants	≥ 96¥	2.5 mL
		Total volume of blood collected for each subject††			**7.8 mL**
	Visit 2 (Day 1)	Blood sample for hematology	All infants	96	1.2 mL
		Total volume of blood collected for each subject			**1.2 mL**
	Visit 4 (Day 7)	Blood sample for hematology	All infants‡	≤96‡	1.2 mL
		~~Blood sample for biochemistry~~	~~All infants~~	~~≤96~~	~~1.1 mL~~
		~~Blood sample for cell-mediated immunogenicity response~~	~~All infants~~	~~96~~	~~2.0 mL~~
		Total volume of blood collected for each subject‡			**~~4.3~~ *1.2* mL**
	Visit 5 (Day 30)	Blood sample for hematology	All infants	96	1.2 mL
		Blood sample for biochemistry	All infants	96	1.1 mL
		Blood sample for cell-mediated immunogenicity response	All infants††	≤96††	2.0 mL
		Blood sample for humoral response	All infants	96	2.5 mL
		Total volume of blood collected for each subject††			**6.8 mL**
	Visit 6 (Day 31)	Blood sample for hematology	All infants	96	1.2 mL
		Total volume of blood collected for each subject			**1.2 mL**
	Visit 8 (Day 37)	Blood sample for hematology	All infants‡	≤96‡	1.2 mL
		~~Blood sample for biochemistry~~	~~All infants~~	~~≤96‡~~	~~1.1 mL~~
		~~Blood sample for cell-mediated immunogenicity response~~	~~All infants~~	~~96~~	~~2.0 mL~~
		Total volume of blood collected for each subject‡			**~~4.3~~ *1.2* mL**
	Visit 9 (Day 60)	Blood sample for hematology	All infants	96	1.2 mL
		Blood sample for biochemistry	All infants	96	1.1 mL
		Blood sample for cell-mediated immunogenicity response	All infants††	≤96††	2.0 mL
		Blood sample for humoral response	All infants	96	2.5 mL
		Total volume of blood collected for each subject††			**6.8 mL**
	Total volume of blood collected for each subject††,‡ from Screening to Visit 9 (Day 60)*				**~~32.4~~ *26.2* mL**
	Visit 10 (Day 365)	Blood sample for cell-mediated immunogenicity response	All infants††	≤96††	2.0 mL
		Blood sample for humoral response	All infants	96	2.5 mL
		Total volume of blood collected for each subject††			**4.5 mL**
	Assessment of potential RSV-RTI	Blood sample for assessment of mechanism of illness (potential ERD)	Hospitalization for RSV-LRTI	Hospitalization for RSV-LRTI	2.5 mL
	Unscheduled visit for safety	Blood sample for hematology	Event-driven	Event-driven	1.2 mL
		Blood sample for biochemistry	Event-driven	Event-driven	1.1 mL
	Total volume of blood collected for each subject††,‡ from Screening to Visit 11 (Day 730)*				**~~36.9~~ *30.7* mL**

Sample	Timepoint	Type of sample †	Subject	Number of subjects	Quantity
Nasal swab	Assessment of potential RSV-RTI	Nasal swab**	Event-driven	Event-driven	-
	Surveillance for asymptomatic RSV-RTI	Nasal swab***	All infants	96	-

** If during passive or active surveillance contact, the investigator/study staff assesses that an infant presents a potential RSV-RTI (refer to Table 4 for definition), the potential RSV infection will be assessed by quantitative ***RT-***PCR of nasal swab specimens taken at an assessment visit.

*** During RSV season, there will be monthly nasal swabs to detect asymptomatic RSV infections. These swabs will be tested by quantitative ***RT-***PCR at sponsor laboratory.

¥ Blood samples for CMI and for humoral immunogenicity may be postponed until after the RSV serostatus is known and eligibility is confirmed up to or on Day 0 (but before vaccination).

† The priority ranking in blood sampling is hematology > biochemistry > humoral response > CMI response. Venipuncture will not be repeated for CMI and humoral response in case of insufficient volume, at any timepoint.

†† In investigational sites with a laboratory in proximity capable to perform WBS necessary for the CMI assay.

‡ Day 7 hematology testing will be performed if any ≥ Grade 1 for platelet decrease is detected on Day 1 after vaccination. Similarly, hematology testing on Day 37 will be performed if any ≥ Grade 1 for platelet decrease is detected on Day 31.

In Section 6.7.3. Laboratory assays, the following changes have been made:

The following laboratory assays are planned:

- Frequency of interleukin 2, 13, and 17 (IL-2, IL-13 and IL-17), cluster of differentiation 40 ligand (CD40L), 41BB, tumor necrosis factor alpha (TNF-α), and IFN-γ secreting CD3+/CD4+ and CD3+/CD8+ T-cells will be determined by ~~intracellular staining (~~ICS~~)~~ assay on whole blood samples (Table 8).
- RTI will be assessed by:
  - Quantitative ***RT-***PCR that is able to discriminate RSV-A and RSV-B subtypes (Table 10).
  - Qualitative multiplex PCR for detection of a panel of ~~17~~ viruses (Table 10).

Table 8 Cell-Mediated Immunity (CMI)

System	Component	Challenge	Method	Unit	Laboratory
Whole blood	IL-2, CD40L, 41BB, TNF-α, IFN-γ, IL-13, IL-17 secreting CD3+/CD4+ and CD3+/CD8+ T-cells	Peptide pools covering the proteins F, N & M2-1	ICS	Events/10E6 cells	GSK Biologicals[1] ***or designated laboratory***

Table 10 Molecular Biology (PCR tests)

System	Component	Kit / Manufacturer	Method	Unit	Laboratory
Nasal swab	RSV A RNA RSV B RNA	In-house	Quantitative ~~real-time~~ ***RT*** PCR	Copies/ml	GSK Biologicals[1] ***or designated laboratory***
Nasal swab	~~RSV~~ ~~Influenza A, including subtypes H1 and H3~~ ~~Influenza A H1N1 2009~~ ~~Influenza B~~ ~~Parainfluenza virus type 1, 2, 3, and 4~~ ~~Human Metapneumovirus~~ ~~Enterovirus/Rhinovirus~~ ~~Adenovirus~~ ~~Bocavirus~~ ~~Coronavirus - 229E, OC43, NL63, HKU1~~ ***Influenza A virus (Flu A)*** ***Influenza B virus (Flu B)*** ***Human respiratory syncytial virus A (RSV A)*** ***Human respiratory syncytial virus B (RSV B)*** ***Human Influenza A virus subtype H1 (Flu A-H1)*** ***Human Influenza A virus subtype H3 (Flu A-H3)*** ***Human Influenza A virus subtype H1pdm09 (Flu A-H1pdm09)*** ***Human adenovirus (AdV)*** ***Human metapneumovirus (MPV)*** ***Human enterovirus (HEV)*** ***Human parainfluenza virus 1 (PIV1)*** ***Human parainfluenza virus 2 (PIV2)*** ***Human parainfluenza virus 3 (PIV3)*** ***Human parainfluenza virus 4 (PIV4)*** ***Human bocavirus 1/2/3/4 (HBoV)*** ***Human rhinovirus A/B/C (HRV)*** ***Human coronavirus 229E (229E)*** ***Human coronavirus NL63 (NL63)*** ***Human coronavirus OC43 (OC43)*** [2]	~~xTAG Respiratory Viral Panel Fast~~ ***Allplex Respiratory Panel or equivalent*** [2]	Multiplex PCR	Qualitative assay (positive/ negative)	GSK Biologicals[1] ***or designated laboratory***

RT: Reverse Transcription;

[2] The list of components might be subject to change in case equivalent kit is used for multiplex RVP testing.

Table 11 Hematology and biochemistry tests

System	Discipline	Component	Timepoint	Method	Scale	Laboratory
Whole blood	Hematology	Hemoglobin	Screening, Day 1, Day 7*, Day 30, Day 31, Day 37*, Day 60	As per local practice	Quantitative	Local laboratory
		Leukocytes (White Blood Cells)				
		Platelets				
Serum	Biochemistry	Alanine Aminotransferase	Screening, ~~Day 7*~~, Day 30, ~~Day 37*~~, Day 60	As per local practice	Quantitative	
		Aspartate Aminotransferase				
		Creatinine				

**** Day 7 hematology testing will be performed if any ≥ Grade 1 for platelet decrease is detected on Day 1 after vaccination. Similarly, hematology testing on Day 37 will be performed if any ≥ Grade 1 for platelet decrease is detected on Day 31***.

In Section 6.7.4.1. Immunological read-outs, the following changes have been made:

Table 12 Immunological read-outs

Blood sampling timepoint			No. subjects	Component	Components priority rank
Type of contact and timepoint	Sampling timepoint	System			
Screening (Day -29 to Day 0)	Pre-Vaccination	Whole blood	~~≤96*,¥~~	IL-2, CD40L, 41BB, TNF-α, IFN-γ, IL-13, IL-17 secreting T-cells	1
		Serum	≥ 96	Respiratory Syncytial virus Ab.IgG (RSV F or G antibody)	1
			~~≥96¥~~	Anti-RSV A Neutralizing Antibody	2
				Respiratory Syncytial Virus F protein Ab.IgG (Anti-RSV PreF antibody)	3
				Respiratory Syncytial Virus F protein Ab.IgG (Palivizumab competing Ab)	4
~~Visit 4 (Day 7)~~	~~Post-Vaccination 1~~	~~Whole blood~~	~~~96~~	~~IL-2, CD40L, 41BB, TNF-α, IFN-γ, IL-13, IL-17 secreting T-cells~~	~~1~~
Visit 5 (Day 30)	Post-Vaccination 1	Whole blood	~~≤96*~~	IL-2, CD40L, 41BB, TNF-α, IFN-γ, IL-13, IL-17 secreting T-cells	1
		Serum	~96	Anti-RSV A Neutralizing Antibody	1
				Respiratory Syncytial Virus F protein Ab.IgG (Anti-RSV PreF antibody)	2
				Respiratory Syncytial Virus F protein Ab.IgG (Palivizumab competing Ab)	3
~~Visit 8 (Day 37)~~	~~Post-Vaccination 2~~	~~Whole blood~~	~~~96~~	~~IL-2, CD40L, 41BB, TNF-α, IFN-γ, IL-13, IL-17 secreting T-cells~~	~~1~~
Visit 9 (Day 60)	Post-Vaccination 2	Whole blood	~~≤96*~~	IL-2, CD40L, 41BB, TNF-α, IFN-γ, IL-13, IL-17 secreting T-cells	1
		Serum	~96	Anti-RSV A Neutralizing Antibody	1
				Respiratory Syncytial Virus F protein Ab.IgG (Anti-RSV PreF antibody)	2
				Respiratory Syncytial Virus F protein Ab.IgG (Palivizumab competing Ab)	3
Visit 10 (Day 365)	Post-Vaccination 2	Whole blood	~~≤96*~~	IL-2, CD40L, 41BB, TNF-α, IFN-γ, IL-13, IL-17 secreting T-cells	1
		Serum	~96	Anti-RSV A Neutralizing Antibody	1
				Respiratory Syncytial Virus F protein Ab.IgG (Anti-RSV PreF antibody)	2

**** Only in investigational sites with a laboratory in proximity capable to perform WBS necessary for the CMI assay.***

† Blood samples for CMI taken on Day 7 and Day 37 for subjects enrolled under protocol amendment 1 will still be assessed for CMI.

¥ Blood samples for CMI and for humoral immunogenicity may be postponed until after the RSV serostatus is known and eligibility is confirmed up to or on Day 0 (but before vaccination).

Table 13 Molecular biology for nasal swab and specimen analysis

Blood sampling timepoint			No. subjects	Component	Components priority rank
Type of contact and timepoint	Sampling timepoint	System			
Surveillance for asymptomatic RSV-RTI	Monthly contact during the RSV season	Nasal swab	~96	Quantitative ~~real-time~~ ***RT-***PCR (RSV A RNA and RSV B RNA)	Not applicable
Assessment of potential RSV-RTI	All subjects attending assessment visit	Nasal swab	Event-driven	Quantitative ~~real-time~~ ***RT-***PCR (RSV A RNA and RSV B RNA)	Not applicable
Assessment of potential RSV-RTI	All subjects attending assessment visit with positive Quantitative RSV A/B ***RT-***PCR results	Nasal swab	Event-driven	Qualitative multiplex PCR** (~~RSV; Influenza A [including subtypes H1 and H3]; Influenza A H1N1 2009; Influenza B; Parainfluenza virus type 1, 2, 3, and 4; Human Metapneumovirus; Enterovirus/Rhinovirus; Adenovirus; Bocavirus; Coronavirus - 229E, OC43, NL63, HKU1~~ ***Influenza A virus (Flu A)*** ***Influenza B virus (Flu B)*** ***Human respiratory syncytial virus A (RSV A)*** ***Human respiratory syncytial virus B (RSV B)*** ***Human Influenza A virus subtype H1 (Flu A-H1)*** ***Human Influenza A virus subtype H3 (Flu A-H3)*** ***Human Influenza A virus subtype H1pdm09 (Flu A-H1pdm09)*** ***Human adenovirus (AdV)*** ***Human metapneumovirus (MPV)*** ***Human enterovirus (HEV)*** ***Human parainfluenza virus 1 (PIV1)*** ***Human parainfluenza virus 2 (PIV2)*** ***Human parainfluenza virus 3 (PIV3)*** ***Human parainfluenza virus 4 (PIV4)*** ***Human bocavirus 1/2/3/4 (HBoV)*** ***Human rhinovirus A/B/C (HRV)*** ***Human coronavirus 229E (229E)*** ***Human coronavirus NL63 (NL63)*** ***Human coronavirus OC43 (OC43)***)	Not applicable
Assessment of potential RSV-RTI	All subjects presenting confirmed LRTI* (with negative Quantitative RSV A and B ***RT-***PCR results)	Nasal swab	Event-driven	Qualitative multiplex PCR** (~~RSV; Influenza A [including subtypes H1 and H3]; Influenza A H1N1 2009; Influenza B; Parainfluenza virus type 1, 2, 3, and 4; Human Metapneumovirus; Enterovirus/Rhinovirus; Adenovirus; Bocavirus; Coronavirus - 229E, OC43, NL63, HKU1~~ ***Influenza A virus (Flu A)*** ***Influenza B virus (Flu B)*** ***Human respiratory syncytial virus A (RSV A)*** ***Human respiratory syncytial virus B (RSV B)*** ***Human Influenza A virus subtype H1 (Flu A-H1)***	Not applicable

Blood sampling timepoint			No. subjects	Component	Components priority rank
Type of contact and timepoint	Sampling timepoint	System			
				Human Influenza A virus subtype H3 (Flu A-H3) ***Human Influenza A virus subtype H1pdm09 (Flu A-H1pdm09)*** ***Human adenovirus (AdV)*** ***Human metapneumovirus (MPV)*** ***Human enterovirus (HEV)*** ***Human parainfluenza virus 1 (PIV1)*** ***Human parainfluenza virus 2 (PIV2)*** ***Human parainfluenza virus 3 (PIV3)*** ***Human parainfluenza virus 4 (PIV4)*** ***Human bocavirus 1/2/3/4 (HBoV)*** ***Human rhinovirus A/B/C (HRV)*** ***Human coronavirus 229E (229E)*** ***Human coronavirus NL63 (NL63)*** ***Human coronavirus OC43 (OC43)***)	

[**] ***The list of components might be subject to change in case equivalent kit is used for multiplex RVP testing.***

In Section 7.2. Storage and handling of study vaccines, the following changes have been made:

For the placebo any temperature excursion outside the range of 0.0 to +8.0°C (for +2 to +8°C/+36 to +46°F label storage condition) and for the ChAd155-RSV vaccine any temperature excursion above -60°C (for ≤ -60°C/≤ -76°F label storage condition) ***or below -90°C*** must be reported in the appropriate (electronic) temperature excursion decision form ([e]TDF).

In Section 7.5. Contraindications to subsequent vaccination, the following changes have been made:

- Any Grade 1 abnormality with potential clinical relevance or any ≥ Grade 2 abnormality in hematological~~/biochemical~~ parameters detected in the Day 7 blood sample (refer to Section 6.6.11.2, Figure 2 and Appendix C).

In Section 9.2.1. Passive surveillance, the following changes have been made:

All subjects' parent(s)/LAR(s) will be instructed to contact investigator/study staff in case of ***any new*** RTI symptoms (cough, runny nose or blocked nose) or in case of ***any new*** difficulty in breathing or wheezing. They will be also reminded to record the start date and the end date of RTI symptoms on the RTI episode card.

- **If there is *any new*** difficulty in breathing, wheezing or parental concern, the investigator/study staff will schedule an assessment visit as soon as possible (refer to Section 9.2.4), but no later than 24 hours after the phone contact to ensure prompt assessment of the need for medical care.
- **If there is <u>no</u> suspicion** of difficulty in breathing, nor wheezing, nor parental concern, but there are ***any new*** symptoms of an RTI (cough, runny nose or blocked nose), the investigator/study staff will schedule an assessment visit as soon as possible (refer to Section 9.2.4), but no later than 72 hours after the phone contact.

In Section 9.2.3. Active surveillance, the following changes have been made:

If there has not been a contact through a clinic visit***/home visit/phone call*** (i.e. Visits 1, 2, 3, 4, 5, 6, 7, 8, 9, 10, 11), a passive surveillance contact (refer to Section 9.2.1), an assessment visit (refer to Section 9.2.4) or a surveillance for asymptomatic RSV infection (refer to Section 9.2.2), then the investigator/study staff will contact the subject's parent(s)/LAR(s).

- Confirm with the subjects' parent(s)/LAR(s), if the subject has developed new RTI symptoms (cough, runny nose or blocked nose) and if he/she has developed any symptoms of difficulty in breathing or wheezing (during and between contacts).
  - **If there is *any new or ongoing*** difficulty in breathing, wheezing or parental concern, the investigator/study staff will schedule an assessment visit as soon as possible (refer to Section 9.2.4), but no later than 24 hours after the contact to ensure prompt assessment of the need for medical care.
  - **If there is <u>no</u> suspicion** of difficulty in breathing, nor wheezing, nor parental concern but ***any new*** symptoms of an RTI (cough, runny nose or blocked nose) are present, the investigator/study staff will schedule an assessment visit as soon as possible (refer to Section 9.2.4), but no later than 72 hours after the contact.

In Section 9.3.1. Time period for detecting and recording adverse events, adverse events of specific interest and serious adverse events, the following changes have been made:

Table 18 Reporting periods for collecting safety information

Visit	SCR	V1	V2	V3		V4		V5	V6	V7		V8		V9	V10	V11
Days	-29 to 0	0	1	3	6	7	29	30	31	33	36	37	59	60	365	730
Clinical examination with special attention given to the detection of episodes of spontaneous bleeding or easy bruising and evidence of bruising or petechiae*																
Examination of limb for petechiae induced by the tourniquet**																

SCR: Screening

* Clinical examination with special attention given to the detection of spontaneous bleeding or easy bruising and evidence of bruising or petechiae will be performed ***by the investigator/study staff*** on Day 1, Day 3, and Day 7 after each vaccination. ***The visits on Day 3 and Day 33 may be replaced by a phone call by the investigator/study staff to inquire of the infants' general health and to solicit reports of a rash, spontaneous bleeding or easy bruising.*** Subjects' parent(s)/LAR(s) will be instructed to contact the investigator/study staff if their child presents symptoms of spontaneous bleeding or easy bruising ***or if their child develops a rash in the month*** following vaccination***, in order to detect any thrombocytopenic petechiae or purpura.***

On Day 7 and/or Day 37, special attention to detection of petechiae or evidence of petechiae induced by the tourniquet will be given in case hematology testing will be performed on that day.

In Section 9.4.2. Contact information for reporting serious adverse events and adverse events of specific interest, the following changes have been made:

Back-up Study Contact for Reporting SAEs and AEs of specific interest
24/24 hour and 7/7 day availability: **GSK Biologicals Clinical Safety & Pharmacovigilance** Outside US ***& Canada sites***: Fax: PPD or PPD Email address: PPD US sites only: Fax: PPD ~~, Tel:~~ PPD ***Canadian sites only:*** ***Fax:*** PPD

In Section 9.10.2. Internal safety review committee (iSRC) oversight, the following changes have been made:

The iSRC will review ***all accumulating*** ~~accumulated~~ safety data three weeks after the start of vaccination and then about every three weeks (until the IDMC ~~reviews the data~~ ***has reviewed all safety data up to 30 days after administration of Dose 2 in Step 3 (i.e. Day 60***).

In Section 9.10.3. Independent Data Monitoring Committee (IDMC) oversight, the following changes have been made:

- The IDMC will receive the following safety data ~~at the end of Steps 1, 2, and 3~~ ***monthly throughout the period of vaccination (up to 30 days after administration of Dose 2 in Step 3, i.e. Day 60):***
- The IDMC will receive the following safety data approximately monthly from beginning of the first RSV season and the frequency will be adapted based on the instruction of the IDMC:
  - Cumulative reports of the incidence of RSV-RTI, RSV-LRTI, ~~and~~ RSV-severe LRTI ***and RSV-RTI leading to hospitalization***.

In Section 9.10.4. Dose-escalation and safety evaluations by iSRC and IDMC, the following changes have been made:

The study will be conducted in a staggered manner to ensure maximum safety of the participating infants ***progressing sequentially through the 3 dose levels*** (Figure 1):

- **Step 1:** vaccination of 32 infants (approximately 16 infants in the RSV-Ld group and approximately 16 infants in the placebo-Ld group). An iSRC will review ***all accumulating*** ~~accumulated~~ safety data three weeks after the start of vaccination in Step 1 and then about every three weeks (until the IDMC ~~reviews~~ ***has reviewed all safety data up to 30 days after administration of Dose 2 in Step 3 (i.e. Day 60).*** ~~the data). As soon as safety data up to 7 days after administration of Dose 2 for all~~

~~infants in Step 1 are available, the entire set of safety data will be reviewed by the IDMC. If there is a favorable outcome of the safety evaluation, enrolment and vaccination (Dose 1) of the infants in Step 2 can start.~~ ***The IDMC will review all accumulating reactogenicity and safety data monthly throughout the period of vaccination (up to 30 days after administration of Dose 2 in Step 3, i.e. Day 60). Dose escalation will proceed after administration of the vaccine to all 32 subjects in the previous Step, and in the absence of a safety concern detected by the IDMC in the regular monitoring of accumulating safety data. It will be ensured that, as a minimum the IDMC will have reviewed safety data of at least 16 subjects for two doses (in Step 1) and given their formal approval prior to enrolling subjects to the next dose level (Step 2).***

- **Step 2:** vaccination of 32 infants (approximately 16 infants in the RSV-Md group and approximately 16 infants in the placebo-Md group). An iSRC will review ***all accumulating*** ~~accumulated~~ safety data three weeks after the start of vaccination in Step 2 and then about every three weeks (until the IDMC ~~reviews~~ ***has reviewed all safety data up to 30 days after administration of Dose 2 in Step 3 (i.e. Day 60).***~~the data). As soon as safety data up to 7 days after administration of Dose 2 for all infants in Step 2 are available, the entire set of safety data will be reviewed by the IDMC. If there is a favorable outcome of the safety evaluation, enrolment and vaccination (Dose 1) of the infants in Step 3 can start.~~ ***The IDMC will review accumulating reactogenicity and safety data monthly throughout the period of vaccination (up to 30 days after administration of Dose 2 in Step 3, i.e. Day 60). Dose escalation will proceed after administration of the vaccine to all 32 subjects in the previous Step, and in the absence of a safety concern detected by the IDMC in the regular monitoring of accumulating safety data. It will be ensured that, as a minimum the IDMC will have reviewed safety data of at least 16 subjects for two doses (in Step 2) and given their formal approval prior to enrolling subjects to the next dose level (Step 3).***
- **Step 3:** vaccination of 32 infants (approximately 16 infants in the RSV-Hd group and approximately 16 infants in the placebo-Hd group). An iSRC will review ***all accumulating*** ~~accumulated~~ safety data three weeks after the start of vaccination in Step 3 and then about every three weeks (until the IDMC ~~reviews~~ ***has reviewed all safety data up to 30 days after administration of Dose 2 in Step 3 (i.e. Day 60).*** A review of the entire set of safety data from Steps 1, 2 and 3 will be performed by the IDMC after availability of safety data up to 30 days after administration of Dose 2 in Step 3 (i.e. Day 60).

In Section 11.1. Primary endpoint, the following changes have been made:

- Occurrence of AEs from first vaccination (Day 0) up to Day 60~~, in all infants~~.
  - Occurrence of episode of spontaneous or excessive bleeding (AE of specific interest), during a 30-day follow-up period after each vaccination~~, in all subjects~~.
  - Occurrence of any biochemical (alanine aminotransferase, aspartate aminotransferase and creatinine) laboratory abnormalities at Screening, ~~Day 7,~~ Day 30, ~~Day 37,~~ and Day 60.

In Section 11.2. Secondary endpoints, the following changes have been made:

- Occurrence of SAEs from study start (Day 0) up to study conclusion (Day 730) ~~in all subjects~~.
- Occurrence of RSV-LRTI (AE of specific interest) as from Dose 1 administration up to study conclusion (Day 730) ~~in all subjects~~.
- Occurrence of RSV-RTI, RSV-LRTI, severe RSV-LRTI (according to ***standardized*** case definitions) as from Dose 1 administration up to study conclusion (Day 730) ~~in all subjects~~.
- Magnitude of the CMI response to the investigational RSV vaccine, pre-vaccination (Screening), post-Dose 1 (~~Day 7 and~~ Day 30) and post-Dose 2 (~~Day 37,~~ Day 60 and Day 365)~~, in all subjects~~**.**
  - CD3+/CD4+ ~~and CD3+/CD8+~~ T-cells expressing ~~at least one marker~~ ***at least two markers among CD40L, IL-2, TNF-α, and IFN-γ upon stimulation with F, N and M2-1 peptide pools***.
- Humoral response to the investigational RSV vaccine, pre-vaccination (Screening), post-Dose 1 (Day 30) and post-Dose 2 (Day 60 and Day 365)~~, in all subjects~~:
  - Neutralizing antibody titers against RSV-A.
  - RSV F antibody concentrations.
- Humoral response to the investigational RSV vaccine, pre-vaccination (Screening), post-Dose 1 (Day 30) and post-Dose 2 (Day 60)~~, in all subjects~~:
  - Palivizumab-competing antibody concentrations.

In Section 11.3. Tertiary endpoints, the following changes have been made:

- CMI response ***profile (Th1, Th2, Th17)*** to the investigational RSV vaccine, pre-vaccination (Screening), post-Dose 1 (~~Day 7 and~~ Day 30) and post-Dose 2 (~~Day 37,~~ Day 60 and Day 365)~~, in all subjects~~**.**
  - CD3+/CD4+ and***/or*** CD3+/CD8+ T-cells expressing ~~any marker or a combination of markers, for determination of Th profile~~ ***at least one or any combination of immune marker(s) among CD40L, 41BB, IL-2, TNF-α, IFN-γ, IL-13 and IL-17 upon stimulation with F, N and M2-1 peptide pools***.
- Any further exploratory immunology to detect ***disease–related or*** vaccine-related immune responses, such as but not limited to:
  - Anti-vector immunity: neutralization.

In Section 11.6.4. RTI and LRTI, the following changes have been made:

For the analysis of RTI and LRTI, all cases will be definitively classified as either RTI, LRTI or severe LRTI according to ~~the~~ ***standardized*** case definitions ~~presented in~~ ***(see*** Table 4***) based on the available WHO case definitions***, and the association to RSV infection will be assessed by quantitative PCR as primary analysis. ~~The analysis may also be done using the available WHO case definitions for RTI, LRTI and severe LRTI.~~

In Section 11.9.1.1. Analysis of secondary objectives, the following changes have been made:

The following parameters will be summarized by group using descriptive statistics, at each timepoint during which blood samples are collected for CMI ***in investigational sites with a laboratory in proximity capable to perform WBS necessary for the CMI assay***:

- ~~Frequency of CD8+ T-cells and T-helper cell profile (Th1, Th2, Th17) based on IL-2, CD40L, 41BB, TNF-α, IFN-γ, IL-13, and IL-17 secreting CD3+/CD4+ and CD3+/CD8+ T-cells.~~
- ***Frequency of CD3+/CD4+ T-cells expressing at least two markers among CD40L, IL-2, TNF-α, and IFN-γ upon stimulation with F, N and M2-1 peptide pools.***

In Section 11.9.1.2. Analysis of tertiary objectives, the following changes have been made:

The cellular immune response will be further characterized using descriptive analysis of the frequency of CD3+/CD8+ T-cells and CD3+/CD4+ T-helper cell profile (Th1, Th2, Th17) expressing at least one or any combination of marker(s) among IL-2, CD40L, 41BB, TNF-α, IFN-γ, IL-13, and IL-17.

In Section 11.10. Analysis of RTI and LRTI, the following changes have been made:

As primary analysis, the assessment of RSV infection will be performed using the quantitative ***RT-***PCR according to ***standardized*** case definitions ~~presented in~~ ***(see*** Table 4) ***based on the available WHO case definitions***. ~~The analysis may also be done using the available WHO case definitions for RTI, LRTI and severe LRTI.~~

Descriptive analyses (mean, median, min, max) of viral load assessed by the quantitative ***RT-***PCR (RSV-A/B) of RSV-RTI, RSV-associated LRTI and severe LRTI cases will be tabulated. This analysis will also be done by study group.

The incidence rate of asymptomatic RSV infections (with 95% CI) detected by the quantitative PCR (RSV-A/B) will be tabulated by group. Descriptive analyses (mean, median, min, max) of viral load assessed by the quantitative ***RT-***PCR (RSV-A/B) of those asymptomatic RSV infections will also be done by group.

In Section 14. References, the following changes have been made:

Modjarrad K, Giersing B, Kaslow DC, Smith PG, Moorthy VS; WHO RSV Vaccine Consultation Expert Group. WHO consultation on Respiratory Syncytial Virus Vaccine Development Report from a World Health Organization Meeting held on 23-24 March 2015. Vaccine. 2016;34(2):190-7.

~~Zent O, Arras-Reiter C, Broeker M *et al*. Immediate allergic reactions after vaccinations-a post-marketing surveillance review. *Eur J Pediatr*. 2002; 161: 21-5.~~

In Appendix A Laboratory assays, the following changes have been made:

Assay descriptions could be subjects to change, due to assay re-development and/or qualification.

Intracellular staining (ICS)

The results are expressed as the frequency of CD4+ or CD8+ T-cells expressing, per million of CD4+ or CD8+ T-cells:

- ***CD3+/CD4+ T-cells expressing at least two markers among CD40L, IL-2, TNF-α, and IFN-γ (poly-functional CD4+ T-cells).*** ~~At least one immune marker (to detect and measure the CD4 or CD8 T-cell response).~~
- ***CD3+/CD4+ and/or CD3+/CD8+ T-cells expressing at least one or any combination of immune marker(s) among CD40L, 41BB, IL-2, TNF-α, IFN-γ, IL-13 and IL-17 (for determination of Th profile).*** ~~Any Th specific immune marker (to determine the Th profile of the CD4 response).~~

PCR

- Qualitative multiplex PCR for detection of a panel of viruses.

  A qualitative PCR multiplex assay is used for the detection and identification of multiple respiratory virus nucleic acids in nasal swabs from individuals suspected of respiratory tract infections. The following virus types and subtypes can be identified in the assay:

  - ***Influenza A virus (Flu A)***
  - ***Influenza B virus (Flu B)***
  - ***Human Influenza A virus subtype H1 (Flu A-H1)***
  - ***Human Influenza A virus subtype H3 (Flu A-H3)***
  - ***Human Influenza A virus subtype H1pdm09 (Flu A-H1pdm09)***
  - ***Human respiratory syncytial virus A (RSV A)***
  - ***Human respiratory syncytial virus B (RSV B)***
  - ***Human adenovirus (AdV)***
  - ***Human metapneumovirus (MPV)***
  - ***Human enterovirus (HEV)***
  - ***Human parainfluenza virus 1 (PIV1)***
  - ***Human parainfluenza virus 2 (PIV2)***
  - ***Human parainfluenza virus 3 (PIV3)***
  - ***Human parainfluenza virus 4 (PIV4)***
  - ***Human bocavirus 1/2/3/4 (HBoV)***

- ***Human rhinovirus A/B/C (HRV)***
- ***Human coronavirus 229E (229E)***
- ***Human coronavirus NL63 (NL63)***
- ***Human coronavirus OC43 (OC43)***

Following total nucleic acids extraction, viruses are detected by multiplex real-time RT-PCR assays targeting the above mentioned viruses. A comparative analysis of the fluorescence intensities of each target is performed to detect the viruses present in the sample.

- ~~17 viruses: xTAG™ RVP FAST assay:~~

  ~~After extraction of nucleic acids from the sample, a single multiplex RT-PCR is performed using random primers. Then, each complementary deoxyribonucleic acid is detected by virus-specific primers, each primer containing a unique tag sequence. A deoxyribonucleic acid polymerase extends the primer when there is a perfect match on the 3' end of the virus specific primer. Biotin-dCTP is incorporated into the extending chain if extension occurs.~~

  ~~Tagged amplified deoxyribonucleic acid is mixed with Luminex beads. These beads are coated with anti-tag oligonucleotides complementary to the tag sequences included in the virus-specific primers. These beads are colored making them spectrally distinguishable from each other. Each tagged primer hybridizes only to its unique anti-tag complement; therefore, each bead color represents a specific virus through the bead/anti-tag/tagged primer association.~~

  ~~Revelation is performed by using a fluorescent reporter molecule (streptavidin-phycoerythrin) that binds to the Biotin-dCTP included in the extended sequences. The beads are then analyzed by the Luminex® 200 instrument which corresponds to a flow-cytometer containing two lasers: one laser allows identification of the color-coded bead associated to each virus and one laser allows identification of the presence or absence of extended primer through the reporter molecule. The data generated by the Luminex® 200 system are expressed in Median Fluorescence Intensity units per bead, which are translated in a positive or negative status for each target.~~

GlaxoSmithKline Biologicals SA

Vaccines R &D

Protocol Amendment 3

eTrack study number and Abbreviated Title:	204838 (RSV PED-002)
IND number:	16999
EudraCT number:	2016-000117-76
Amendment number:	Amendment 3
Amendment date:	12 September 2017
Co-ordinating authors:	• PPD (Scientific Writer, Keyrus Biopharma contractor for GSK Biologicals) • PPD, Scientific Writer

Rationale/background for changes:

- The Spanish competent authorities (Agencia Española de Medicamentos y Productos Sanitarios [AEMPS]) requested the Sponsor to add the inclusion criterion of being born full-term, which was removed for Protocol Amendment 2. This inclusion criterion in Section 5.2 will be required for Spain.
- The collection of birth weight and gestation at birth in weeks at the screening visit has been added to the list of study procedures (Table 5) in Section 6.5 and described in Section 6.6.3. This procedure will be applicable to all participating countries and will permit the identification of pre-term infants recruited in the study.

Amended text has been included in *bold italics* and deleted text in ~~strikethrough~~ in the following sections:

On **the protocol cover page**, the following change has been made:

Co-ordinating authors • PPD ***, Scientific Writer***

In Section 5.2. Inclusion criteria, the following change has been made:

- ***Born full-term (i.e. after a gestation period of 37 to less than 42 completed weeks) with a minimum birth weight of 2.5 kg. (Required for Spain***)

In Section 6.5. Outline of study procedures, the following change has been made:

Table 5 List of study procedures

Epoch	Epoch 001										Epoch 002					
Age	12-23 mth										24-35 mth	36-47 mth				
Type of contact	Screening	Visit 1	Visit 2#	Visit 3##	Visit 4#	Visit 5	Visit 6#	Visit 7##	Visit 8#	Visit 9	Visit 10	Visit 11#	Unscheduled visit for safety*	Contact for active/passive surveillance**	Surveillance for asymptomatic RSV-RTI***	Assessment of potential RSV-RTI****
Timepoints	D-29 to D0	D0	D1	D3	D7	D30	D31	D33	D37	D60	D365	D730		Monthly or Weekly	Monthly	
Collect birth weight and gestation at birth in weeks	•															

In Section 6.6.3. Collect demographic data, the following change has been made:

Record demographic data such as date of birth (day, month and year), gender, geographic ancestry, and ethnicity in the subject's eCRF. ***Collect and record the birth weight and gestation at birth (in weeks) in the eCRF.***

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Vaccines R &D

Protocol Amendment 4

eTrack study number and Abbreviated Title:	204838 (RSV PED-002)
IND number:	16999
EudraCT number:	2016-000117-76
Amendment number:	Amendment 4
Amendment date:	10 December 2017
Co-ordinating authors:	• PPD , Scientific Writer

Rationale/background for changes:

- An Independent Data Monitoring Committee (IDMC) is reviewing all accumulating unblinded safety and reactogenicity data on a monthly basis for this study to ensure that there is a timely identification of any safety signal. As safety data accumulates, it may be that there is sufficient evidence of safety of the current dose level to allow progression to the next dose level. For instance, taking the *a priori* safety concern of thrombocytopenia, this amendment will apply both Frequentist and Bayesian approaches to the existing data, as described in Section 11.4 to show the likelihood of observing more extreme values. The number of subjects evaluated by the IDMC for the two-step dose escalation to steps 2 and 3 after administration of two doses of study vaccine may continue to be 32 subjects at steps 1 and 2 as before. However, in the absence of a significant safety concern detected in the regular monitoring of all parameters of accumulating safety data, the IDMC agreed to recommend that dose escalation could potentially proceed on at least 16 subjects, without requiring the enrolment and evaluation of the full group size of 32 subjects. These amendment changes were implemented in the Synopsis and Synopsis Table 1, as well as Sections 1.2.4.1, 1.2.4.2, 1.3, Figure 1 in Section 3, Table 1 in Section 3, Sections 5.1, 6.2.2.2.1, Table 7 in 6.7.2, Table 11 in Section 6.7.3, Table 12 and Table 13 in Section 6.7.4.1, and in Sections 9.10.4 and 11.4.
- A footnote was added to indicate the requirement to record the white blood cell differential including absolute neutrophil and lymphocyte counts in the eCRF to Table 11 in Section 6.7.3 and the toxicity grading scales for the absolute neutrophil and lymphocyte counts were indicated in Table 25 of Appendix C.
- In addition, some typographical errors have been corrected throughout the protocol.

Amended text has been included in *bold italics* and deleted text in ~~strikethrough~~ in the following sections:

On the protocol cover page, the following changes were made:

Contributing authors

- PPD ***(Project Statistician)***
- PPD ***, (Clinical Trial Supply Manager)***

- PPD *(Central Safety Physician)*
- PPD *(Safety Scientist)*

In the **Synopsis**, the following changes have been made:

Rationale for the study and study design

Rationale for the study design

- Study blinding: Given the different ~~appearance and~~ storage conditions of the investigational RSV vaccine and placebo, double blinding is not possible and the study will be conducted in an observer-blind manner.

  The investigational ChAd155-RSV vaccine will be administered to ***up to*** 48 subjects in total. As a control, ***up to*** 48 subjects will be vaccinated with placebo across the three steps.

  – In **Step 1**, ~~32~~***between a maximum of*** 32 ***to a minimum of 16*** RSV-seropositive infants will receive two doses of either low dose ChAd155-RSV vaccine (5×10^9 vp [RSV-Ld]; 0.5 mL) or placebo (0.5 mL), according to their random assignment.

  – In **Step 2**, ***between a maximum of*** 32 ***to a minimum of 16*** RSV-seropositive infants will receive two doses of either middle dose ChAd155-RSV vaccine (1.5×10^{10} vp [RSV-Md]; 0.15 mL) or placebo (0.15 mL), according to their random assignment.

  The iSRC and IDMC will determine whether any of the predefined study holding rules are met. If this is the case, vaccination in the study will be immediately put on hold. At any time, the IDMC may recommend to the sponsor to suspend the enrolment to the study and/or vaccination based on the safety data regularly arising in this study.

  Dose escalation will proceed after administration of the vaccine to all ~~32~~ subjects in the previous ~~Step, and in the absence~~ of a ***step if no significant*** safety concern ***is*** detected by the IDMC in ~~the~~***their*** regular ~~monitoring~~***review*** of accumulating safety data. ~~It will be ensured that, as a minimum, the~~***The*** IDMC ~~will have reviewed~~***may recommend for dose escalation to proceed in the absence of a concern based on*** safety data ***review on a minimum*** of ~~at least~~ 16 subjects ~~for two doses and given their formal approval prior to enrolling subjects~~ ***completing dose 2. The IDMC may recommend that based on the evolving safety profile, sufficient information has been accumulated to allow safe progression*** to the next ***sequential*** dose level***. Thus, the number of subjects enrolled to the low and mid dose is between 16 and 32 subjects but the number of***

subjects enrolled to the high dose remains fixed at 32 subjects.

In the **Synopsis Table 1 Study groups and epochs foreseen in the study a footnote** was added as follows:

The target will be to enroll up to 96 RSV-seropositive infants aged 12 to 23 months at the time of first vaccination (unless the dose escalation IDMC decision can be made based on the evaluation of accumulating safety data on fewer subjects) to ensure that at least 64 infants receive two doses of study vaccine (ChAd155-RSV or Placebo). Then the number of subjects at each step will be at least 16 (approximately 8 subjects per group) at steps 1 and 2 and 32 (approximately 16 subjects per group) at step 3.

Number of subjects	The target will be to enroll ~~approximately~~ ***up to*** 96 RSV-seropositive infants ~~(32 infants per step)~~ aged 12 to 23 months at the time of first vaccination ***(unless the dose escalation IDMC decision can be made based on the evaluation of accumulating safety data on fewer subjects)*** to ensure that ~~96~~***at least 64*** infants receive ~~at least one dose~~ ***two*** dose***s*** of study vaccine. ***(ChAd155-RSV or Placebo)***.

LIST OF ABBREVIATIONS

mm³	***cubic millimeter***

In Section 1.2.4. Rationale for safety monitoring plan, the following changes have been made:

The Section 1.2.4.1. Rationale for earlier dose escalation decision making, was added.

Under Amendment 4 of the protocol, the IDMC may on reviewing of the accumulating safety data recommend that dose escalation can safely proceed based on the experience of a minimum of 16 subjects.

Taking the a priori safety concern of thrombocytopenia, both Frequentist and Bayesian approaches are applied to provide complementary assurance on the possibility to exclude a signal based on the accumulated safety data. The Frequentist approach examines the statistical power to rule out a significant decrease in platelet counts due to vaccination that would suggest a four-fold or greater increase in the risk of a Grade 3 or higher thrombocytopenia with 16 subjects per dose cohort. The Bayesian approach calculates the posterior predictive probability of not observing any subject with a Grade 3 or higher thrombocytopenia among the future 16 subjects by basing it on what we have observed in the first 16 subjects (see Section 11.4).

In Section 1.2.4.2 Staggered design with 3 steps, the following changes have been made:

The target will be to enroll ~~approximately~~ ***up to*** 96 RSV-seropositive infants ***aged 12 to 23 months at the time of first vaccination (unless the dose escalation IDMC decision can be made based on the evaluation of accumulating safety data on fewer subjects)*** to ensure that ~~96~~***at least 64*** infants receive ~~at least one dose~~ ***two*** dose***s*** of study vaccine~~. The~~ ***(***ChAd155-RSV ~~vaccine will be administered to 48 subjects in total. As a control, 48 subjects will be administered with placebo across the three steps.~~***or Placebo)***.

- In **Step 1**, ~~32~~***between a maximum of*** 32 ***to a minimum of 16*** RSV-seropositive infants will receive two doses of either low dose ChAd155-RSV vaccine (5 x 10^9 vp [RSV-Ld]; 0.5 mL) or placebo (0.5 mL), according to their random assignment.
- In **Step 2**, ***between a maximum of*** 32 ***to a minimum of 16*** RSV-seropositive infants will receive two doses of either middle dose ChAd155-RSV vaccine (1.5 x 10^{10} vp [RSV-Md]; 0.15 mL) or placebo (0.15 mL), according to their random assignment.
- In **Step 3,** 32 RSV-seropositive infants will receive two doses of either high dose ChAd155-RSV vaccine (5 x 10^{10} vp [RSV-Hd]; 0.5 mL) or placebo (0.5 mL), according to their random assignment.

Dose escalation will proceed after administration of ***two doses of*** the vaccine to ~~all~~ 32 subjects ~~in the previous Step, and~~ . ***However,*** in the absence of a ***significant*** safety concern detected in the regular monitoring of accumulating safety data ***on at least 16 subjects, the IDMC may allow for dose escalation to proceed without the requirement of enrolling and evaluating the full group size of 32 subjects. A subsequent dose escalation will proceed after administration of two doses of the vaccine to between a maximum of 32 to a minimum of 16 subjects in in the absence of a significant*** safety concern detected by the IDMC in the regular monitoring of accumulating safety data. ***Thus, upon review of the accumulating safety data on a minimum of 16 subjects after administration of two doses of the vaccine, the IDMC may decide that based on the evolving safety profile, sufficient information has been accumulated to allow safe progression to the next sequential dose level.*** It will be ensured that, as a minimum the IDMC will have reviewed safety data of at least 16 subjects for two doses and given their formal approval prior to enrolling subjects to the next dose level.

In Section 1.2.5. Rationale for study blinding, the following change has been made:

Given the different ~~appearance and~~ storage conditions of the investigational RSV vaccine and placebo, double blinding is not possible and the study will be conducted in an observer-blind manner.

In Section 1.3. Benefit : Risk Assessment, the following change has been made:

Approximately ~~H~~***h***alf of the infants (***between 32 and*** 48) in this study will be exposed to the ChAd155-RSV vaccine, whereas the other infants will receive a placebo.

In Section 3. STUDY DESIGN OVERVIEW, the following change has been made:

In the **Figure 1 Study Design,** footnote d has been modified as follows:

d Dose escalation will proceed after administration of ***two doses of*** the vaccine to ~~all~~ 32 subjects in ~~the previous~~ Step~~,~~ ~~and~~ ***1***. ***However,*** in the absence of a ***significant*** safety concern detected in the regular monitoring of accumulating safety data ***on at least 16 subjects, the IDMC may allow for dose escalation to proceed without the requirement of enrolling and evaluating the full group size of 32 subjects. A subsequent dose escalation (from Step 2 to Step 3) will proceed after administration of two doses of the vaccine to between a maximum of 32 to a minimum of 16 subjects in the absence of a significant*** safety concern detected by the IDMC in the regular monitoring of accumulating safety data***. Thus, upon review of the accumulating safety data on a minimum of 16 subjects after administration of two doses of the vaccine, the IDMC may decide that based on the evolving safety profile, sufficient information has been accumulated to allow safe progression to the next sequential dose level***. It will be ensured that, as a minimum the IDMC will have reviewed safety data of at least 16 subjects for two doses and given their formal approval prior to enrolling subjects to the next dose level.

In the **Table 1 Study groups and epochs foreseen in the study** a footnote was added as follows:

****The target will be to enroll up to 96 RSV-seropositive infants aged 12 to 23 months at the time of first vaccination (unless the dose escalation IDMC decision can be made based on the evaluation of accumulating safety data on fewer subjects) to ensure that at least 64 infants receive two doses of study vaccine (ChAd155-RSV or Placebo) (see Section 5.1). Then the number of subjects at each step will be at least 16 (approximately 8 subjects per group) at steps 1 and 2 and 32 (approximately 16 subjects per group at step 3).***

In Section 5.1. Number of subjects, the following change has been made:

The target will be to enroll ~~approximately~~ ***up to*** 96 RSV-seropositive infants ~~(32 infants per step)~~ aged 12 to 23 months at the time of first vaccination ***(unless the dose escalation IDMC decision can be made based on the evaluation of accumulating safety data on fewer subjects*** to ensure that ~~96~~***at least 64*** infants receive ~~at least one dose~~ ***two*** dose***s*** of study vaccine ***(ChAd155-RSV or Placebo)***. Refer to Section 11.4 for the determination of sample size.

In Section 6.2.2.2.1. Study group and treatment number allocation, the following changes have been made:

The target will be to enroll ~~approximately~~ ***up to*** 96 RSV-seropositive infants ~~(32 infants per step)~~aged 12 to 23 months at the time of first vaccination ***(unless the dose escalation IDMC decision can be made based on the evaluation of accumulating safety data on fewer subjects*** to ensure that ~~96~~***at least 64*** infants receive ~~at least one dose~~ ***two*** dose***s*** of study vaccine ***(ChAd155-RSV or Placebo) (see Section 5.1)***. Infants will be randomly assigned to two study groups per step in a (1:1) ratio (approximately 16 subjects in each group).

- In **Step 1**, ***between*** approximately ***8 and*** 16 RSV-seropositive infants will be randomly assigned to receive low dose ChAd155-RSV vaccine (5×10^9 vp [RSV-Ld]; 0.5 mL) and ***between*** approximately ***8 and*** 16 RSV-seropositive infants will be randomly assigned to receive placebo (0.5 mL).
- In **Step 2**, ***between*** approximately ***8 and*** 16 RSV-seropositive infants will be randomly assigned to receive middle dose ChAd155-RSV vaccine (1.5×10^{10} vp

[RSV-Md]; 0.15 mL) and ***between*** approximately ***8 and*** 16 RSV-seropositive infants will be randomly assigned to receive placebo (0.15 mL).

- In **Step 3,** approximately 16 RSV-seropositive infants will be randomly assigned to receive high dose ChAd155-RSV vaccine (5 x 10^{10} vp [RSV-Hd]; 0.5 mL) and approximately 16 RSV-seropositive infants will be randomly assigned to receive placebo (0.5 mL).

In Section 6.6.8. Assess pre-vaccination body temperature, the following change has been made:

The axillary, rectal, oral, or tympanic body temperature of all subjects needs to be measured prior to any study vaccines administration.

In Section 6.7.2. Biological samples, the following changes have been made:

In the **Table 7 Biological samples**, a footnote was added as follows:

Sample	Timepoint	Type of sample†	Subject	Number of subjects§	Quantity

§ The target will be to enroll up to 96 RSV-seropositive infants aged 12 to 23 months at the time of first vaccination (unless the dose escalation IDMC decision can be made based on the evaluation of accumulating safety data on fewer subjects) to ensure that at least 64 infants receive two doses of study vaccine (ChAd155-RSV or Placebo) (see Section 5.1).

In Section 6.7.3. Laboratory assays, the following changes have been made:

In the **Table 11 Hematology and biochemistry tests**, a footnote was added as follows:

System	Discipline	Component	Timepoint	Method	Scale	Laboratory
Whole blood	Hematology	Hemoglobin	Screening, Day 1, Day 7*, Day 30, Day 31, Day 37*, Day 60	As per local practice	Quantitative	Local laboratory
		Leukocytes (White Blood Cells)**				
		Platelets				
Serum	Biochemistry	Alanine Aminotransferase	Screening, Day 30, Day 60	As per local practice	Quantitative	
		Aspartate Aminotransferase				
		Creatinine				

***** White Blood Cell differential including absolute neutrophil and lymphocyte counts must be recorded in the eCRF.***

In Section 6.7.4.1. Immunological read-outs, the following changes have been made:

In the **Table 12 Immunological read-outs**, a footnote was added as follows:

Blood sampling timepoint			No. subjects†	Component	Components priority rank
Type of contact and timepoint*	**Sampling timepoint**	**System**			
Screening (Day -29 to Day 0)	Pre-Vaccination	Whole blood	≤96*,¥,‡,¥	IL-2, CD40L, 41BB, TNF-α, IFN-γ, IL-13, IL-17 secreting T-cells	1
		Serum	≥ 96	Respiratory Syncytial virus Ab.IgG (RSV F or G antibody)	1
			≥96¥	Anti-RSV A Neutralizing Antibody	2
				Respiratory Syncytial Virus F protein Ab.IgG (Anti-RSV PreF antibody)	3
				Respiratory Syncytial Virus F protein Ab.IgG (Palivizumab competing Ab)	4
Visit 5 (Day 30)	Post-Vaccination 1	Whole blood	≤96*‡	IL-2, CD40L, 41BB, TNF-α, IFN-γ, IL-13, IL-17 secreting T-cells	1
		Serum	~96	Anti-RSV A Neutralizing Antibody	1
				Respiratory Syncytial Virus F protein Ab.IgG (Anti-RSV PreF antibody)	2
				Respiratory Syncytial Virus F protein Ab.IgG (Palivizumab competing Ab)	3
Visit 9 (Day 60)	Post-Vaccination 2	Whole blood	≤96*‡	IL-2, CD40L, 41BB, TNF-α, IFN-γ, IL-13, IL-17 secreting T-cells	1
		Serum	~96	Anti-RSV A Neutralizing Antibody	1
				Respiratory Syncytial Virus F protein Ab.IgG (Anti-RSV PreF antibody)	2
				Respiratory Syncytial Virus F protein Ab.IgG (Palivizumab competing Ab)	3
Visit 10 (Day 365)	Post-Vaccination 2	Whole blood	≤96*‡	IL-2, CD40L, 41BB, TNF-α, IFN-γ, IL-13, IL-17 secreting T-cells	1
		Serum	~96	Anti-RSV A Neutralizing Antibody	1
				Respiratory Syncytial Virus F protein Ab.IgG (Anti-RSV PreF antibody)	2

CD40L: cluster of differentiation 40 ligand; **IFN-γ**: interferon gamma; **IgG**: immunoglobulin G; **IL (IL-2; IL-13; IL-17)**: interleukin; **TNF-α**: tumor necrosis factor alpha; **RSV**: respiratory syncytial virus.

~~* Only in investigational sites with a laboratory in proximity capable to perform WBS necessary for the CMI assay.~~

‡* Blood samples for CMI taken on Day 7 and Day 37 for subjects enrolled under protocol amendment 1 will still be assessed for CMI.

† The target will be to enroll up to 96 RSV-seropositive infants aged 12 to 23 months at the time of first vaccination (unless the dose escalation IDMC decision can be made based on the evaluation of accumulating safety data on fewer subjects) to ensure that at least 64 infants receive two doses of study vaccine (ChAd155-RSV or Placebo) (see Section 5.1).

‡ Only in investigational sites with a laboratory in proximity capable to perform WBS necessary for the CMI assay.

¥ Blood samples for CMI and for humoral immunogenicity may be postponed until after the RSV serostatus is known and eligibility is confirmed up to or on Day 0(but before vaccination).

In the **Table 13 Molecular biology for nasal swab and specimen analysis**, a footnote was added as follows:

Blood sampling timepoint			No. subjects†	Component	Components priority rank
Type of contact and timepoint	**Sampling timepoint**	**System**			

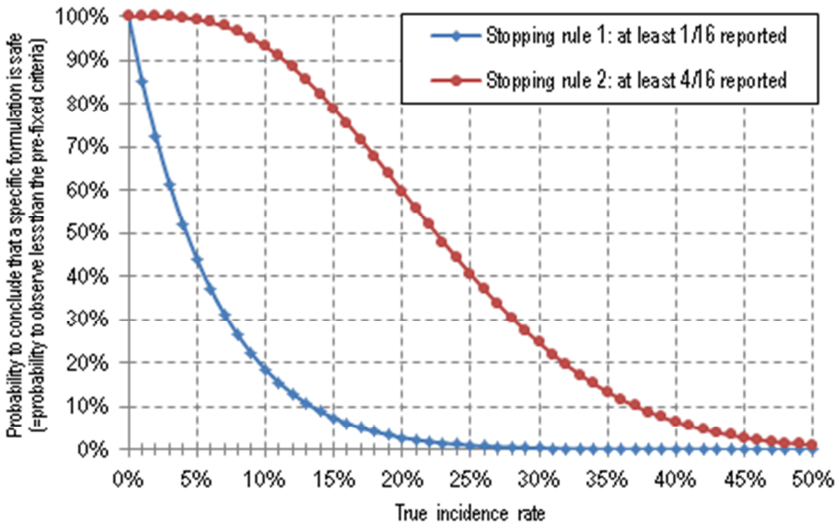
† The target will be to enroll up to 96 RSV-seropositive infants aged 12 to 23 months at the time of first vaccination (unless the dose escalation IDMC decision can be made based on the evaluation of accumulating safety data on fewer subjects) to ensure that at least 64 infants receive two doses of study vaccine (ChAd155-RSV or Placebo) (see Section 5.1).

In Section 9.10.4. Dose-escalation and safety evaluations by iSRC and IDMC, the following changes have been made:

- **Step 1**: vaccination of ***a maximum of*** 32 infants (approximately 16 infants in the RSV-Ld group and approximately 16 infants in the placebo-Ld group). An iSRC will review all accumulating safety data three weeks after the start of vaccination in Step 1 and then about every three weeks (until the IDMC has reviewed all safety data up to 30 days after administration of Dose 2 in Step 3 (i.e. Day 60). The IDMC will review all accumulating reactogenicity and safety data monthly throughout the period of vaccination (up to 30 days after administration of Dose 2 in Step 3, i.e. Day 60). Dose escalation ***to Step 2*** will proceed after ~~administration~~***completion of*** ~~the vaccine to all~~***vaccination f****or* 32 subjects ~~in the previous step~~, and in the absence of a ***significant*** safety concern detected by the IDMC in the regular monitoring of accumulating safety data. ***However, in the absence of a significant safety concern detected in the regular monitoring of accumulating safety data on at least 16 subjects, the IDMC may recommend for dose escalation to proceed without the requirement of enrolling and evaluating the full group size of 32 subjects. A subsequent dose escalation will proceed after administration of two doses of the vaccine to between a maximum of 32 to a minimum of 16 subjects in the absence of a significant safety concern detected by the IDMC in the regular monitoring of accumulating safety data.*** It will be ensured that, as a minimum the IDMC will have reviewed safety data of at least 16 subjects for two doses (in Step 1) and given their formal approval prior to enrolling subjects to the next dose level (Step 2).
- **Step 2**: vaccination of ***a maximum of*** 32 infants (approximately 16 infants in the RSV-Md group and approximately 16 infants in the placebo-Md group). An iSRC will review all accumulating safety data three weeks after the start of vaccination in Step 2 and then about every three weeks (until the IDMC has reviewed all safety data up to 30 days after administration of Dose 2 in Step 3 (i.e. Day 60). The IDMC will review accumulating reactogenicity and safety data monthly throughout the period of vaccination (up to 30 days after administration of Dose 2 in Step 3, i.e. Day 60). Dose escalation ***to Step 3*** will proceed after administration ***of two doses*** of the vaccine to ~~all~~ 32 subjects ~~in the previous step~~, and in the absence of a ***significant*** safety concern detected by the IDMC in the regular monitoring of accumulating safety data. ***However, as for the dose escalation from Step 1 to Step 2, in the absence of a significant safety concern detected in the regular monitoring of accumulating safety data on at least 16 subjects, the IDMC may again allow for dose escalation to proceed without the requirement of enrolling and evaluating the full group size of 32 subjects. A subsequent dose escalation will proceed after administration of two doses of the vaccine to between a maximum of 32 to a minimum of 16 subjects in the absence of a significant safety concern detected by the IDMC in the regular monitoring of accumulating safety data.*** It will be ensured that, as a minimum the IDMC will have reviewed safety data of at least 16 subjects for two doses (in Step 2) and given their formal approval prior to enrolling subjects to the next dose level (Step 3).

In Section 9.10.5. Holding rules, the following change has been made:

Figure 4 Evaluations based on 16 subjects - Risk assessment curve for one formulation based on the proposed safety holding rules



The above figure illustrates that, with ***a maximum of*** 16 subjects per study group:

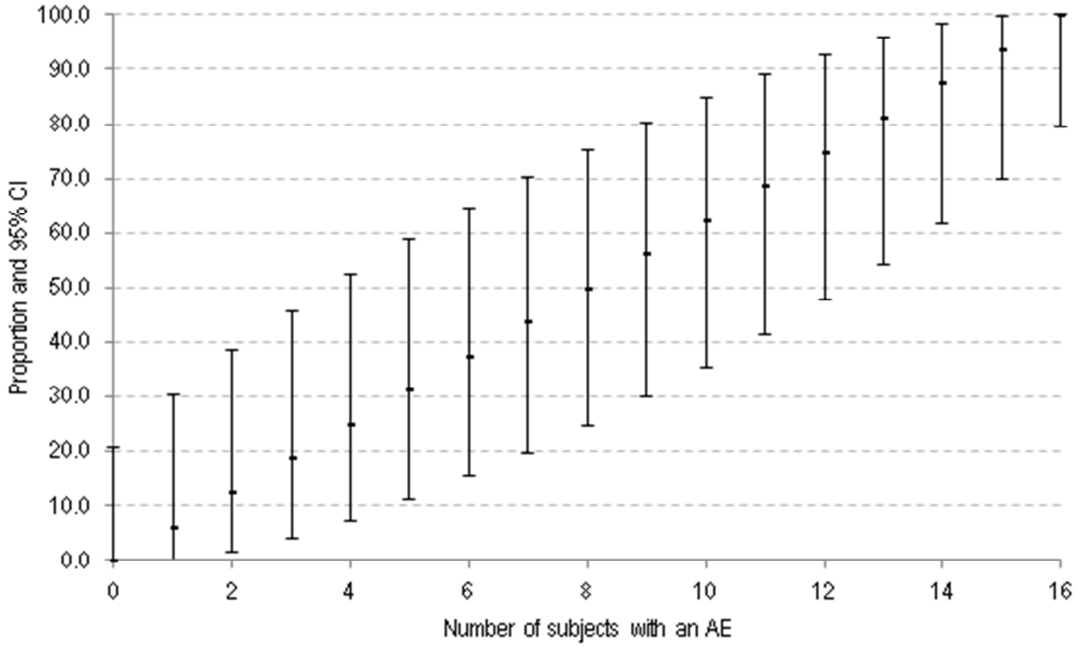
In Section 11.4. Determination of sample size, the following changes have been made:

No hypothesis driven sample size calculation was conducted due to the nature of descriptive summary of the study objectives. The sample size of 32 subjects per dose cohort/Step, randomized 1:1 to vaccine or placebo, was based on conventional study designs for Phase 1/2 trials. Specifically, if no adverse event was observed among 16 RSV vaccinated subjects, the exact, two-sided 95% confidence interval (CI) would rule out an AE rate of 20% or more; if the incidence rate of AEs is more than 10%, there will still be more than an 80% chance of observing at least one subject with this AE. Combining all RSV vaccinees across all dose levels, there will be more than a 99% chance of observing at least one subject with this AE.

Furthermore, with 16 RSV vaccinees, the maximum width of a 95% CI for the proportion of subjects with adverse events among 16 vaccinees is about 50% (see Figure 5).

~~Considering the sample size of 16 subjects in each study group,~~ Figure 5 illustrates the precision on the percentage of subjects with symptoms following vaccination.

Figure 5 Exact 95% confidence interval on the percentage of subjects with adverse events following vaccination for 16 subjects per group



Taking the a priori safety concern of thrombocytopenia, both Frequentist and Bayesian approaches will be applied here to provide complementary assurance on the possibility to exclude a signal based on the accumulated safety data. The Frequentist approach examines the statistical power to rule out a significant decrease in platelet counts due to vaccination that would suggest a four-fold or greater increase in the risk of a Grade 3 or higher thrombocytopenia with 16 subjects per dose cohort. The Bayesian approach calculates the posterior predictive probability of not observing any subject with a Grade 3 or higher thrombocytopenia among the future 16 subjects by basing it on what we have observed in the first 16 subjects.

If we assume platelet counts in healthy infants between 12 and 23 months follow a normal distribution with a mean of 300,000 platelets/mm³ and a standard deviation of 75,000 platelets/mm³, then we would expect 0.04% of unvaccinated infants to have a count below 50000 platelets/mm³, which is the cut-off for a Grade 3 thrombocytopenia. To rule out a four-fold or greater increase, we would need to show that the mean platelet count does not fall below 190,000 platelets/mm³ with a standard deviation of 47,500. The sample size needed to rule out a 110,000 platelets/mm³ decrease due to vaccination with 84% power when vaccination has no effect on platelet count is 16 infants per dose cohort (8 per group). Sample size is based on a one-sided 0.025 test of non-inferiority using the Mann-Whitney statistic (PASS 12, Non-inferiority Tests for Two Means using Differences).

The Bayesian approach to predict the probability of no Grade 3 or higher thrombocytopenia in the future 16 subjects per dose cohort will be conducted by treating the mean of platelet counts μ as a random variable with a normal prior distribution $N(\mu_0, \sigma_0^2)$, where μ_0 and σ_0 will be determined by using laboratory normal

range. For this analysis the range of 150,000 to 450,000 platelets/mm³ will be the focus. Based on this laboratory range, μ_0 would be 300,000, a mean of the range, and σ_0 would be 75000, an approximate of a standard deviation of data sampled from this normal range. After 16 infants complete the two-dose vaccination schedule, a posterior distribution of μ will be calculated based on the prior of μ and the observed platelet counts from Day 31. The posterior predictive probability of no Grade 3 or higher thrombocytopenia in the future 16 subjects per dose cohort will be calculated, and sensitivity on the prior will be assessed.

After the first 16 infants of a dose cohort have completed the two-dose vaccination schedule, the 95% CI for the difference in mean platelets at Day 31 between vaccinees and placebo recipients will be calculated along with the posterior predictive probability of no Grade 3 or higher thrombocytopenia in the future 16 infants. If the lower boundary of the 95% CI is above 110,000 platelets/mm³ and the predictive probability is sufficiently high (e.g., 80% or more), the IDMC may recommend enrolling infants into the next dose cohort without completing enrolment in the current cohort. However, if a Grade 3 or higher thrombocytopenia occurs in a vaccinated infant at any time point or if there is a significant safety concern with another laboratory parameter such as hemoglobin or neutrophils, the IDMC may recommend continuing enrolment into the lower dose cohort.

In Section 11.8.1. Within groups assessment, the following change was made:

The percentage of subjects with at least one **local AE** (solicited and unsolicited), with at least one **general AE** (solicited and unsolicited) and with any AE during the 7-day or 30-day follow-up period will be tabulated with exact 95% ~~confidence interval (~~CI~~)~~ after each vaccine dose and overall.

In Section APPENDIX A LABORATORY ASSAYS, the following changes have been made:

ELISA

- Anti-RSV protein F ELISA

  The anti-F protein IgG ELISA is an indirect ELISA allowing the detection and the quantitation of specific IgG antibodies directed against the RSV F protein in human serum samples.

  ~~First, F protein antigens purified from CHO expression system are coated onto 96-well microplates. Then, after a washing and a blocking step, serial two-fold dilutions of test sera, controls, and reference standard are incubated to allow specific binding of antibodies directed against the F protein antigens. Bound IgG are detected by addition of a goat anti-human IgG antibody conjugated to horseradish peroxidase. After a washing step, the horseradish peroxidase substrate solution (TMB/H_2O_2) is added and a colored product develops proportionally to the amount of anti-F protein IgG antibodies present in the test serum. The color is quantified by reading the optical densities at 450-620 nm using a spectrophotometer. Antibody concentrations of individual serum and control samples are determined after interpolation from the~~

~~reference standard curve using a four-parameter equation and are expressed in arbitrary ELISA units (EU)/mL.~~

In APPENDIX C TOXICITY GRADING FOR HEMATOLOGY AND BIOCHEMISTRY PARAMETERS, the following changes have been made:

In the **Table 25 Toxicity grading scales for hematology and biochemistry parameters applicable for this study**, two grading scales were added as follows:

Component	Grade 1	Grade 2	Grade 3	Grade 4
Hemoglobin (g/dL)	9.0 to < 10.5	8.0 to < 9.0	7.0 to < 8.0	< 7.0
Leukocytes decreased (cell/mm^3)	2500 to < 3500	1500 to < 2500	1000 to < 1500	< 1000
Absolute neutrophil count decreased (cell/mm^3)	***1000 to < 1300***	***750 to < 1000***	***500 to < 750***	***< 500***
Absolute lymphocyte count decreased (cell/mm^3)	***600 to < 650***	***500 to < 600***	***350 to < 500***	***< 350***
Platelets decreased (cell~~/~~/mm^3)	75000 to < 150000	50000 to < 75000	25000 to < 50000	< 25000
Alanine Aminotransferase (increase by factor)	1.1 to < 2.0 xULN	2.0 to < 3.0 xULN	3.00 to ≤ 8.0 xULN	> 8.0 xULN
Aspartate Aminotransferase (increase by factor)	1.1 to < 2.0 xULN	2.0 to < 3.0 xULN	3.00 to ≤ 8.0 xULN	> 8.0 xULN
Creatinine (mg/dL)	0.6 to < 0.9	0.9 to < 1.2	1.2 to ≤ 1.5	> 1.5

Grading scale adapted from [Division of AIDS, 2003], [Division of AIDS, 2004] and [Division of AIDS, 2007].
ULN: upper limit of normal