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

Primary Study vaccines and numbers	GlaxoSmithKline (GSK) Biologicals non-typeable *Haemophilus influenzae* (NTHi) and *Moraxella catarrhalis* (Mcat) multi-antigen vaccine consisting of three conserved surface proteins (PD, PE and PilA) from *Haemophilus influenzae* and one conserved surface protein (UspA2) from Mcat (GSK3277511A): • GSK3277513A (10 µg of PD, 10 µg of PE-PilA and 10 µg of UspA2/ plain or $AS01_E$) • GSK3339036A (10µg of PD, 10 µg of PE-PilA, and 3.3µg of UspA2/ $AS01_E$)
Other Study vaccine	• Control: saline Placebo
eTrack study number and Abbreviated Title	201281 (NTHI MCAT-001)
Investigational New Drug (IND) number	***16531***
EudraCT number	2015-000378-36
Date of protocol	Final Version 1: 09 March 2015 Final Version 2: 30 March 2015 Final Version 3: 22 April 2015
Date of protocol amendment	Amendment 1 Final: 30 July 2015 Amendment 2 Final 16 March 2016
Title	An observer-blind study to evaluate the safety, reactogenicity and immunogenicity of the investigational GSK Biologicals' GSK3277511A vaccine in adults.
Detailed Title	A Phase I, randomised, observer-blind, placebo-controlled, multi-centre study to evaluate the safety, reactogenicity and immunogenicity of GSK Biologicals' GSK3277511A investigational vaccine when administered intramuscularly according to a 0, 2 months schedule in adults.
Co-ordinating author	PPD (XPE Pharma & Science for GSK Biologicals)
Contributing authors	• PPD, Director, Clinical Research & Translational Science

eTrack study number and Abbreviated Title	201281 (NTHI MCAT-001)
Investigational New Drug (IND) number	***16531***
EudraCT number	2015-000378-36
Detailed Title	A Phase I, randomised, observer-blind, placebo-controlled, multi-centre study to evaluate the safety, reactogenicity and immunogenicity of GSK Biologicals' GSK3277511A investigational vaccine when administered intramuscularly according to a 0, 2 months schedule in adults.
Contributing authors (continued) (Amended 16 March 2016)	• PPD, ***Clinical Research and Development Lead*** • PPD, PPD, Lead Biostatistician • PPD, Project Biostatistician • PPD, ***Project Delivery Lead*** • PPD, Study Delivery Lead • PPD (Novellas Healthcare for GSK Biologicals)/PPD, Study Delivery Manager • PPD, ***CLS Clinical Readout Team Leader*** • PPD, ***CLS Laboratory*** Study Manager • PPD /PPD, Clinical Safety Representatives • PPD (Aixial Pharma for GSK Biologicals), Study Data Manager • PPD, Global Regulatory Lead • PPD (CVO-Europe for GSK Biologicals), Vaccine Supply Coordinator

GSK Biologicals' Protocol DS v 14.1.1

Protocol Amendment 2 Sponsor Signatory Approval

eTrack study number and Abbreviated Title	201281 (NTHI MCAT-001)
IND number	***16531***
EudraCT number	2015-000378-36
Date of protocol amendment	Amendment 2 Final 16 March 2016
Detailed Title	A Phase I, randomised, observer-blind, placebo-controlled, multi-centre study to evaluate the safety, reactogenicity and immunogenicity of GSK Biologicals' GSK3277511A investigational vaccine when administered intramuscularly according to a 0, 2 months schedule in adults.
Sponsor signatory (Amended 16 March 2016)	***Ashwani Kumar Arora, Clinical and Epidemiology Project Lead***
Signature	_____
Date	_____

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Protocol Amendment 2 Rationale

Amendment number: Amendment 2
Rationale/background for changes:

- The spirometry test must be of good quality. If this is not the case, a re-test should be done. This re-test is covered by the current Informed Consent Form.
- In compliance with ICH requirements, the protocol mentions that all results will be presented in an integrated report at the end of the study.
- A description of how subjects will be included in the CMI sub-cohort and a description of how the sub-cohort will represent the fully randomized study groups has been added.
- Following re-development and re-validation of the anti-PD ELISA, a new cut-off was defined.
- Additional administrative changes have been performed:
  - The IND number has been added.
  - The list of contributing authors as well as Sponsor signatory has been updated.
  - GSK Biologicals' "Global Vaccines Clinical Laboratories (GVCL)" has been renamed as "Clinical Laboratory Sciences (CLS)"
  - Name "Quest Diagnostic Limited" has been replaced by "Q^2 Solutions" due to its merge with QLab.
  - Reference to the GSK Biologicals' Laval laboratory was removed, as this laboratory will not be used in the study. In addition, this laboratory is no longer part of GSK Biologicals' laboratories.
  - In addition, minor edits in other sections were made for clarification.

Protocol Amendment 2 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 vaccines 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 vaccines, 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	201281 (NTHI MCAT-001)
IND number	***16531***
EudraCT number	2015-000378-36
Date of protocol amendment	Amendment 2 Final: 16 March 2016
Detailed Title	A Phase I, randomised, observer-blind, placebo-controlled, multi-centre study to evaluate the safety, reactogenicity and immunogenicity of GSK Biologicals' GSK3277511A investigational vaccine when administered intramuscularly according to a 0, 2 months schedule in adults.
Investigator name	_____
Signature	_____
Date	_____

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Sponsor Information

1. Sponsor

GlaxoSmithKline Biologicals
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 8.4.2.

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 8.8.

SYNOPSIS

Detailed Title	A Phase I, randomised, observer-blind, placebo-controlled, multi-centre study to evaluate the safety, reactogenicity and immunogenicity of GSK Biologicals' GSK3277511A investigational vaccine when administered intramuscularly according to a 0, 2 months schedule in adults.
Indication	Immunisation against non-typeable *Haemophilus influenzae* (NTHi) and *Moraxella catarrhalis* (Mcat) in adults.
Rationale for the study and study design	• **Rationale for the study** GSK Biologicals is developing a new NTHi-Mcat investigational vaccine against diseases caused by NTHi and Mcat, such as respiratory tract infections in COPD (chronic obstructive pulmonary disease) patients, which can lead to acute exacerbations of COPD (AECOPD). The investigational vaccine that will be evaluated in the present study is a multi-component vaccine based on conserved surface proteins of NTHi and Mcat. Three NTHi proteins have been selected: Protein D (PD), as a free recombinant protein and Protein E (PE) and PilA protein, as a recombinant fusion protein named PE-PilA. The selected Mcat antigen is UspA2. The purpose of this new Phase I study is to evaluate the safety, reactogenicity and immunogenicity of the NTHi-Mcat investigational vaccines, first in healthy young adults (18-40 years old) with a non-adjuvanted formulation and secondly in adults 50–70 years old having a significant smoking history (≥ 10 pack-years) with two adjuvanted formulations. The investigational vaccines will be administered according to a 0, 2 months schedule. Saline placebo will be used as control. • **Rationale for the study design** As the UspA2 antigen has never been administered to human, healthy young adults will be enrolled and vaccinated with a non-adjuvanted vaccine in the study prior to vaccinate adults of 50-70 years old with the adjuvanted formulations. The study will hence follow a staggered design with two steps. Step 1 will include healthy adults 18–40 years old either receiving a non-adjuvanted (plain) vaccine containing 10 µg of PD, 10 µg of PE-PilA and 10 µg of UspA2, or a placebo control. Step 2 will include adults 50–70 years old with a smoking history of at least 10 pack years either receiving placebo control, or one of two $AS01_E$-adjuvanted formulations, i.e. 10 µg of PD, 10 µg of PE-PilA and 10 µg of UspA2 or 10 µg of PD, 10 µg of PE-PilA and 3.3 µg of UspA2.

Moving to Step 2 or administration of vaccine Dose 2 within a step will be conditional on the favourable outcome of an evaluation of safety data obtained up to 7 days after administration of (a) previous vaccine dose(s) by an internal Safety Review Committee (iSRC).

- **Rationale for the use of placebo**

The placebo group (saline solution) is included in the NTHI MCAT-001 study as a control for both the safety and immunogenicity assessments.

Objectives

Primary

- To evaluate the safety and reactogenicity profile of the NTHi-Mcat investigational vaccines.

Secondary

- To evaluate the humoral and cellular immune response of the NTHi-Mcat investigational vaccines.

Tertiary

- To collect blood samples for assay development/validation and/or for evaluation/characterisation of the humoral and cellular immune responses to components of either the NTHi-Mcat investigational vaccines and/or of other respiratory pathogens.

Study design

- **Experimental design:** Phase I, observer-blind, randomised, placebo-controlled, multi-centric study with two steps.
- **Duration of the study:** For each subject, the study will last approximately 15 months, from screening to study end.
  - Epoch 001: Primary starting at Screening Visit and ending at, and including Visit 6 (Day 90).
  - Epoch 002: Follow-up starting at Visit 7 (Day 210) and ending at Visit 8 (Day 420).
- **Study groups:**
  - **10-10-10**: Approximately 15 subjects receiving two doses of the non-adjuvanted GSK Biologicals' NTHi-Mcat investigational vaccine containing 10 µg of PD, PE-PilA and UspA2 during Step 1 of the study.
  - **PLACE1**: Approximately 15 subjects receiving two

doses of placebo (saline solution) during Step 1 of the study.

– **10-10-10-AS:** Approximately 30 subjects receiving two doses of the $AS01_E$-adjuvanted GSK Biologicals' NTHi-Mcat investigational vaccine containing 10 µg of PD, PE-PilA and UspA2 during Step 2 of the study.

– **10-10-3-AS**: Approximately 30 subjects receiving two doses of the $AS01_E$-adjuvanted GSK Biologicals' NTHi-Mcat investigational vaccine containing 10 µg of PD, 10 µg of PE-PilA, and 3.3 µg of UspA2 during Step 2 of the study.

– **PLACE2**: Approximately 30 subjects receiving two doses of placebo (saline solution) during Step 2 of the study.

Synopsis Table 1 Study groups and epochs foreseen in the study

Study groups	Number of subjects	Age (Min/Max)	Epochs		Step
			Epoch 001	Epoch 002	
10-10-10	15	18 - 40 years	x	x	1
PLACE1	15	18 - 40 years	x	x	1
10-10-10-AS	30	50 - 70 years	x	x	2
10-10-3-AS	30	50 - 70 years	x	x	2
PLACE2	30	50 - 70 years	x	x	2

Synopsis Table 2 Study groups and treatment foreseen in the study

Treatment name	Vaccine/ product name	Study Groups				
		10-10-10	10-10-10-AS	10-10-3-AS	PLACE1	PLACE2
10-10-10/Plain	NTHi-Mcat 10-10-10	X				
	NaCl					
10-10-10/$AS01_E$	NTHi-Mcat 10-10-10		X			
	AS01E					
10-10-3/$AS01_E$	NTHi-Mcat 30-30-10 *			X		
	AS01E					
Placebo	NaCl				X	X

* Vials containing 37.5 µg, 37.5 µg and 12.5 µg of freeze-dried PD, PE-PilA and UspA2, respectively, will be diluted in three volumes of $AS01_E$ (total of ~1875 µL). 0.5 mL will be administered.

- **Control:** placebo control (saline solution).
- **Vaccination schedule:** 0, 2 months (i.e. at Visit 1 [Day 0] and Visit 4 [Day 60]).
- **Treatment allocation:** Subjects will be allocated to a study group using a centralised randomisation system on internet (SBIR). SBIR will also be used for treatment allocation at second dose. The randomisation algorithm

will use a minimisation procedure accounting for centre at Step 1 and accounting for age (50-59 years or 60-70 years), for smoking status (current or former smokers), for centre and for forced expiratory volume in 1 second (FEV_1) / forced vital capacity (FVC) (≥ 0.7 or < 0.7) at Step 2.

- **Blinding:** Observer-blind.

Synopsis Table 3 Blinding of study epochs

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

- **Sampling schedule:**
  - Blood samples for safety (haematology/ biochemistry) will be collected from all subjects at Screening Visit (pre-Day 0), at Visit 1 (Day 0), Visit 2 (Day 7), Visit 4 (Day 60), Visit 5 (Day 67), Visit 7 (Day 210) and at Visit 8 (Day 420).
  - Blood samples for immunogenicity will be collected from all subjects for humoral immunity at Visit 1 (Day 0), Visit 3 (Day 30), Visit 4 (Day 60), Visit 6 (Day 90), Visit 7 (Day 210) and at Visit 8 (Day 420), and from a sub-cohort of subjects for cell-mediated immunity (CMI) at Visit 1 (Day 0), Visit 4 (Day 60), Visit 6 (Day 90), Visit 7 (Day 210) and at Visit 8 (Day 420).
- **Type of study:** self-contained
- **Data collection:** Electronic Case Report Form (eCRF)
- **Safety monitoring during vaccination period:** Safety evaluations by the Safety Review Team (SRT) and iSRC.

  Moving to Step 2 or administration of vaccine Dose 2 within a step will depend on the favourable outcome of a safety evaluation based on the safety data collected up to 7 days after administration of the previous dose(s). In order to move to Step 2, safety data will be needed from all subjects. In order to move from Dose 1 to Dose 2 within a step, safety data from about 50% of the vaccinated subjects will be needed.

Number of subjects The target is to enrol ~30 eligible subjects 18–40 years old in Step 1 (~15/group) and ~90 eligible subjects 50–70 years old in Step 2 (~30/group). The total target number of subjects is ~120.

Endpoints

Primary

- Occurrence of each solicited local and general adverse event (AE), during a 7-day follow-up period (i.e. day of vaccination and 6 subsequent days) post-Dose 1 and post-Dose 2, in all subjects, in all vaccine groups.
- Occurrence of any unsolicited AEs, during a 30-day follow-up period (i.e. day of vaccination and 29 subsequent days) post-Dose 1 and post-Dose 2, in all subjects, in all vaccine groups.
- Occurrence of haematological and biochemical laboratory abnormalities, after vaccination, in all subjects, in all vaccine groups:
  - Any haematological (RBC, WBC and differential count, platelets count and haemoglobin level) or biochemical (alanine aminotransferase [ALT], aspartate aminotransferase [AST] and creatinine) laboratory abnormality on Day 7, Day 60, Day 67, Day 210 and Day 420.
- Occurrence of any serious AE (SAE), occurring from first vaccination to study conclusion in all subjects, in all vaccine groups.
- Occurrence of any potential immune-mediated disease (pIMD) occurring from first vaccination to study conclusion in all subjects, in all vaccine groups.

Secondary

- Humoral immune response to the components of the NTHi-Mcat investigational vaccine formulations, on Day 0, Day 30, Day 60, Day 90, Day 210 and Day 420, in all subjects, in all vaccine groups:
  - Anti-PD, anti-PE, anti-PilA and anti-UspA2 antibody concentrations.
- Cell-mediated immune response to components of the NTHi-Mcat investigational vaccine formulations, on Day 0, Day 60, Day 90, Day 210 and Day 420, in a sub-cohort of subjects, in all vaccine groups:
  - Frequency of specific $CD4^+/CD8^+$ T-cells measured on cryopreserved peripheral blood mononuclear cells (PBMCs) and identified by flow cytometry intracellular cytokine staining (ICS) expressing two or more markers (such as IL-2, IL-13, IL-17, IFN-γ, TNF-α and CD40L).

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APPENDIX D AMENDMENTS AND ADMINISTRATIVE CHANGES TO THE PROTOCOL 111

LIST OF ABBREVIATIONS

ANCOVA	analysis of covariance
AE	adverse event
AECOPD	acute exacerbation of COPD
Al	aluminium
ALT	alanine aminotransferase
AS	adjuvant system
AST	aspartate aminotransferase
ATP	according-to-protocol
ATS	American Thoracic Society
CI	confidence interval
***CLS* (Amended 16 March 2016)**	***Clinical Laboratory Sciences***
CMI	cell-mediated immunity
COPD	chronic obstructive pulmonary disease
CRDL	Clinical Research & Development Lead
CSR	clinical study report
eCRF	electronic Case Report Form
ELISA	enzyme-linked immunosorbent assay
EL.U/mL	ELISA unit per millilitre
EMA	European Medicines Agency
ERS	European Respiratory Society
(e)TDF	(electronic) temperature excursion decision form
FDA	Food and Drug Administration, United States
FEV_1	forced expiratory volume in 1 second
FTIH	first time in human

FVC	forced vital capacity
GCP	Good Clinical Practice
GMC	geometric mean concentration
GOLD	Global Initiative for Chronic Obstructive Lung Disease
GSK	GlaxoSmithKline
IB	Investigator Brochure
ICF	Informed Consent Form
ICH	International Conference on Harmonisation
ICS	intracellular cytokine staining
IEC	Independent Ethics Committee
IFN-γ	interferon gamma
Ig	immunoglobulin
IL	interleukin
IM	intramuscular
IMP	investigational medicinal product
IRB	Institutional Review Board
iSRC	internal Safety Review Committee
Mcat	*Moraxella catarrhalis*
MedDRA	medical dictionary for regulatory activities
NaCl	sodium chloride (saline solution)
NTHi	non-typeable *Haemophilus influenzae*
PBMC	peripheral blood mononuclear cell
PD	protein D of *Haemophilus influenzae*
PE	protein E of *Haemophilus influenzae*
PilA	type IV pili subunit of non-typeable *Haemophilus influenzae*

pIMD	potential immune-mediated disease
PT	preferred term
RBC	red blood cells
SAE	serious adverse event
SBIR	randomisation system on internet
SDV	source document verification
SPM	Study Procedures Manual
SRT	Safety Review Team
TNF-α	tumour necrosis factor alpha
TVC	total vaccinated cohort
UspA2	ubiquitous surface protein A2 of *Moraxella catarrhalis*
VSMB	Vaccine Safety Monitoring Board
WBC	white blood cells
WHO	World Health Organisation
µL	microliter

GLOSSARY OF TERMS

Adequate contraception

Adequate contraception is defined as:

1. a contraceptive method with failure rate of less than 1% per year when used consistently and correctly and when applicable, in accordance with the product label for example:

- abstinence from penile-vaginal intercourse, when this is their preferred and usual lifestyle,
- oral contraceptives, either combined or progestogen alone,
- injectable progestogen,
- implants of etenogestrel or levonorgestrel,
- estrogenic vaginal ring,
- percutaneous contraceptive patches,
- intrauterine device or intrauterine system,
- current tubal ligation,
- male partner sterilisation prior to the female subject's entry into the study, and this male is the sole partner for that subject,

  The information on the male sterility can come from the site personnel's review of the subject's medical records; or interview with the subject on her medical history.

2. a contraceptive method with failure rate of more than 1% per year but still considered as acceptable birth control method:

- male condom combined with a vaginal spermicide (foam, gel, film, cream or suppository),
- male condom combined with a female diaphragm, either with or without a vaginal spermicide (foam, gel, film, cream, or suppository).

Adequate contraception does not apply to subjects of child bearing potential with same sex partners, when this is their preferred and usual lifestyle.

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 unfavourable 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.
Alcoholism	Alcoholism, also known as dependency on alcohol or alcohol addiction, is a chronic disease. The signs and symptoms of alcoholism include: • A strong craving for alcohol. • Continued use despite repeated physical, psychological, or interpersonal problems. The inability to limit drinking.
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 (SAE). 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 5.3 for details on observer-blinded studies).
Current smoker	A person who is currently smoking or who has stopped smoking within the past 6 months.
Eligible	Qualified for enrolment into the study based upon strict adherence to inclusion/exclusion criteria.
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 6.6.2 and 10.4 for details on criteria for evaluability).
Former smoker	A person who stopped smoking for at least 6 months.
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.
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 authorisation 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.
Menarche	Menarche is the onset of menses for the first time in a young female and is preceded by several changes associated with puberty including breast development and pubic hair growth. Menarche usually occurs within 1-2 years of breast development, thelarche. However, a young female can become pregnant before her first menses. Thus, a conservative definition of non-childbearing potential in a pre-menarcheal female is a young female who has not yet entered puberty as evidenced by lack of breast development (palpable glandular breast tissue).
Menopause	Menopause is the age associated with complete cessation of menstrual cycles, menses, and implies the loss of reproductive potential by ovarian failure. A practical definition accepts menopause after 1 year without menses with an appropriate clinical profile at the appropriate age e.g. > 45 years.
Pack-years of smoking	Pack-years is a quantification of cigarette smoking, a way to measure the total amount a person has smoked in the course of his/ her lifetime. The number of pack-years is calculated as follows: (average number of *cigarettes* smoked per day x number of years smoked)/ 20 E.g. a smoking history of 10 pack-years means having smoked 20 cigarettes per day for 10 years, or having smoked 10 cigarettes per day for 20 years. *Note*: For the purpose of this study, pipe and/or cigar use

	should not be used to calculate pack-year history.
Potential immune-mediated disease	Potential immune-mediated diseases (pIMDs) are a subset of AEs that include autoimmune diseases and other inflammatory and/or neurologic disorders of interest which may or may not have an autoimmune aetiology.
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 Harmonisation (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.
Randomisation	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 adverse event	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.
Sub-cohort	A group of subjects for whom specific study procedures are planned as compared to other subjects.
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 vaccines 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 products or marketed product(s) or placebo intended to be administered to a subject, identified by a unique number, according to the study randomisation or treatment allocation.
Treatment number	A number identifying a treatment to a subject, according to the study randomisation or treatment allocation.
Unsolicited adverse event	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 adverse event.

1. INTRODUCTION

1.1. Background

1.1.1. COPD: an introduction

Chronic Obstructive Pulmonary Disease (COPD), a common preventable disease, is characterised by persistent airflow limitation that is usually progressive and associated with an enhanced chronic inflammatory response in the airways and lungs to noxious particles of gases. The most important environmental risk factor for COPD is tobacco smoking, even though other factors, such as occupational exposure, may also contribute to the development of the disease [GOLD, 2014]. It is a multi-component disease with several contributory mechanisms, including airway inflammation, airway obstruction, mucociliary dysfunction and structural changes to the airways [American Thoracic Society and European Respiratory Society, 2004; GOLD, 2014]. Together, these mechanisms manifest as an accelerated decline in lung function, with symptoms such as breathlessness on physical exertion, deteriorating health status and exacerbations.

The prevalence of COPD is increasing. Worldwide, COPD (GOLD grade II and above) affects 10.1±4.8% of the population ≥40 years of age [Buist, 2007]. COPD is most prevalent in adults/elderly with a history of smoking [Mannino, 2002]. It is the fourth leading cause of chronic morbidity and mortality in the United States and the first in terms of disease burden in China. According to the World Health Organisation (WHO), COPD is expected to be the third cause of death worldwide by 2020 [Rabe, 2007].

Acute exacerbations and comorbidities contribute to the overall disease severity in individual COPD patients. An acute exacerbation of COPD (AECOPD) is defined as an acute event characterised by a worsening of the patient's respiratory symptoms that is beyond normal day-to-day variations and leads to a change in medication [GOLD, 2014]. The rate at which AECOPD occur varies greatly between patients, with severe patients exacerbating more often. AECOPD additionally increase morbidity and mortality, lead to faster decline in lung function, poorer functional status, and have a significant impact on healthcare systems worldwide [Sapey, 2006]. Between 40-60% of medical expenditure for COPD is a direct consequence of AECOPD [Cazzola, 2008].

The lungs are known to be colonised with different strains of bacteria [Erb-Downward, 2011]. In COPD patients, acquisition of new bacterial strains is believed to be an important cause of AECOPD [Sethi, 2002]. Although estimates vary widely, non-typeable *Haemophilus influenzae* (NTHi) appears to be the main bacterial pathogen associated with AECOPD (11-38%), followed by *Moraxella catarrhalis* (Mcat) (3-25%) and *Streptococcus pneumoniae* (4-9%) [Alamoudi, 2007; Bandi, 2003; Hutchinson, 2007; Ko, 2007; Larsen, 2009; Murphy, 2005; Papi, 2006; Rosell, 2005; Sethi, 2002; Sethi, 2008; Wilkinson, 2006].

1.1.2. Current management of AECOPD

A wide range of pharmacologic (such as inhaled corticosteroids, bronchodilators, phosphodiesterase inhibitors, theophyllines, long-term antibiotics and mucolytics) and

non-pharmacologic (such as lung volume reduction surgery, home oxygen, ventilatory support and pulmonary rehabilitation) interventions exist to manage or treat COPD, some with a positive impact on the AECOPD rate. However, a need for further novel interventions remains because current approaches are not completely effective, even when targeted and used optimally.

Prevention of AECOPD is an insufficiently addressed medical need today, despite existing preventative therapies (bronchodilators such as long-acting muscarinic antagonists, long-acting beta agonists, methylxanthines, corticosteroids, phosphodiesterase-4 inhibitors and combination drugs), and is thought to remain so in the 10 years horizon.

The use of antibiotics is recommended by several guidelines [American Thoracic Society and European Respiratory Society, 2004] as a standard treatment for COPD patients with AECOPD showing purulent sputum. However, as not all patients have confirmed bacterial-related exacerbations, there is an inappropriate use of antibiotics, leading to the spread of antibiotic-resistant bacteria [Daubin, 2008]. Infections with multidrug-resistant bacteria have been linked to increases in morbidity, length of hospitalisation, health care cost and mortality [Nseir, 2008].

There is currently no vaccine indicated for prevention of AECOPD, even though influenza and pneumococcal vaccines are routinely recommended to COPD patients. The availability of a vaccine for the prevention of bacterial AECOPD could contribute significantly to the current management of COPD, in terms of reducing the risk of bacterial exacerbations as well as the inappropriate use of antibiotics.

1.1.3. GSK Biologicals' NTHi-Mcat investigational vaccine

GlaxoSmithKline (GSK) Biologicals is developing a new NTHi-Mcat investigational vaccine against diseases caused by NTHi and Mcat. The investigational vaccine that will be evaluated in the present study is an adjuvanted multi-component vaccine consisting of conserved surface proteins from NTHi and Mcat. Three NTHi proteins have been selected: Protein D (PD), as a free recombinant protein and Protein E (PE) and PilA protein, as a recombinant fusion protein named PE-PilA. The selected Mcat antigen is the UspA2.

PD acts as a glycerophosphodiesterase and is involved in the adhesion and invasion via phosphorylcholine decoration of bacterial membrane's components. Passive transfer of anti-PD sera induces protection in the NTHi chinchilla otitis media model [Novotny, 2006], as well as vaccination with PD induces protection in the mouse nasopharyngeal colonisation model [Forsgren, 2008].

During the infection process, PE is involved in the adhesion of the bacteria to human respiratory and mucosal cells and in the resistance to human complement, a key component of the innate immune system. PilA is involved in bacterial biofilm formation and in the adhesion to human cells mechanism. Anti-PilA antibodies were shown to strongly reduce biofilm formation, *in vitro* and *in vivo* in preclinical animal models [Jurcisek, 2007]. It was demonstrated that antibodies against PE and PilA strongly reduces nasopharyngeal colonisation in mice and/or otitis media in a chinchilla model

[Ronander, 2009; Jurcisek, 2007; Novotny, 2009; *GSK unpublished data*]. Preclinical experiments demonstrated that even if PE and PilA are expressed as a fusion protein (PE-PilA), the biological activities of elicited antibodies against PE and PilA are kept [*GSK unpublished data*].

UspA2 is a putative autotransporter macromolecule and is a vitronectin-binding protein. UspA2 seems to be involved in the ability of Mcat to resist to the bactericidal activity of normal human serum [Attia, 2005]. UspA2 has been shown to induce cross-bactericidal antibodies production and cross-protection in a mouse lung colonisation model [Chen, 1996].

The target population of GSK NTHi-Mcat vaccine are COPD patients, which are often elderly, who are known to have a weakened immune response due to functional defects and altered frequencies of innate and adaptive immune cells, with impaired generation of long-term immune memory (immunosenescence) [Weinberger, 2008]. Moreover, the immune response of COPD patients has been suggested to be disturbed both locally and systemically. Using an adjuvant is thought to help to induce a higher and longer-lasting immune response.

Several formulations of a vaccine containing the NTHi antigens (10 or 30 µg) either non-adjuvanted or combined with different adjuvants (aluminium [Al], adjuvant system [AS]01_E and $AS04_C$) were already evaluated in two previous Phase I clinical trials (NTHI-002 in healthy adults 18-40 years old and NTHI-003 in current and former healthy smokers 50-70 years old). The investigational vaccines were well-tolerated, with an acceptable safety and reactogenicity profile. These studies allowed the dose selection of the NTHi antigens (i.e. 10 µg) and the adjuvant system (i.e. $AS01_E$) to be used in the current study.

Refer to the current Investigator Brochure (IB) for information regarding the completed pre-clinical and clinical (NTHi) studies of the NTHi-Mcat investigational vaccine.

1.2. Rationale for the study and study design

1.2.1. Rationale for the study

The purpose of this new Phase I study is to evaluate the safety, reactogenicity and immunogenicity of the NTHi-Mcat investigational vaccine, first in healthy young adults (18-40 years old) with a non-adjuvanted formulation and secondly in adults 50–70 years old having a significant smoking history (≥ 10 pack-years) with two adjuvanted formulations. The investigational vaccines will be administered according to a 0, 2 months schedule. Saline placebo will be used as control.

As the prevalence of COPD increases with age and as age has an influence on both the immunogenicity and reactogenicity of a vaccine, subjects 50–70 years old will be enrolled in Step 2 of this study. As cigarette smoking is the most commonly encountered risk factor for COPD, adults with a smoking history of at least 10 pack-years have been selected in order to immunologically match the COPD population as much as possible. Literature data indeed suggest that alterations of the immune system start early on in

smokers, before the COPD disease is recognised [Barcelo, 2008; Droemann, 2005; Takanashi, 1999].

1.2.2. Rationale for the study design

As the UspA2 antigen has never been administered to human, healthy young adults will be enrolled and vaccinated with a non-adjuvanted vaccine prior to vaccinate adults 50-70 years old with adjuvanted formulations. The study will hence follow a staggered design with two steps:

- **Step 1** will include 30 healthy adults 18–40 years old either receiving a non-adjuvanted (plain) vaccine containing 10 µg of PD, 10µg of PE-PilA and 10µg of UspA2, or a placebo control (15/group).
- **Step 2** will include 90 healthy adults 50–70 years old with a smoking history of at least 10 pack-years, either receiving placebo control, or one of two $AS01_E$-adjuvanted formulations, i.e. 10 µg of PD, 10 µg of PE-PilA and 10 µg of UspA2 (Group 10-10-10-AS) or 10 µg of PD, 10 µg of PE-PilA and 3.3 µg of UspA2 (Group 10-10-3-AS) (30/group).

Moving to Step 2 or administration of vaccine Dose 2 within a step will be conditional on the favourable outcome of an evaluation of safety data obtained up to 7 days after administration of (a) previous vaccine dose(s) by an internal Safety Review Committee (iSRC).

1.2.3. Rationale for the use of placebo

The placebo group (saline solution) is included in the NTHI MCAT-001 study as a control for both safety and immunogenicity assessments.

1.3. Benefit/Risk Assessment

Please refer to the current IB for the summary of potential risks and benefits of the NTHi-Mcat investigational vaccine.

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

1.3.1. Risk Assessment

Important Potential/Identified Risk	Data/Rationale for Risk	Mitigation Strategy
NTHi-Mcat investigational vaccine		
Theoretical risk of acquiring a vaccine-induced autoimmune disease after vaccination	No confirmed signals related to this potential risk have been identified during the preclinical and clinical programs so far (data from two studies using NTHi vaccines: NTHI-002 (non-adjuvanted formulations) and NTHI-003 (adjuvanted and non-adjuvanted formulations).	Close monitoring of potential immune-mediated diseases in clinical development programs using adjuvants systems. The potential risk of events of possible autoimmune aetiology to occur is mentioned in the Informed Consent Form (ICF).

1.3.2. Benefit Assessment

Benefits considerations include:

- Contribution to the process of developing of a vaccine against AECOPD.
- Medical evaluations/assessments associated with this study (i.e. physical examination, blood testing [haematology and biochemistry data], spirometry [only in Step 2]).
- Early detection of COPD in heavy smokers or elderly.
- Subjects with mild COPD/pulmonary obstruction may benefit from the vaccine (i.e. possible prevention of AECOPD caused by NTHi and/or Mcat).

1.3.3. Overall Benefit/Risk Conclusion

Taking into account the measures taken to minimise risks to subjects participating in this study, the potential and/or known risks identified in association with the candidate NTHi-Mcat vaccines are justified by the anticipated benefits that may be afforded to patients for the prevention of AECOPD.

2. OBJECTIVES

2.1. Primary objective

- To evaluate the safety and reactogenicity profile of the NTHi-Mcat investigational vaccines.

Refer to Section 10.1 for the definition of the primary endpoints.

2.2. Secondary objective

- To evaluate the humoral and cellular immune response of the NTHi-Mcat investigational vaccines.

Refer to Section 10.2 for the definition of the secondary endpoints.

2.3. Tertiary objective

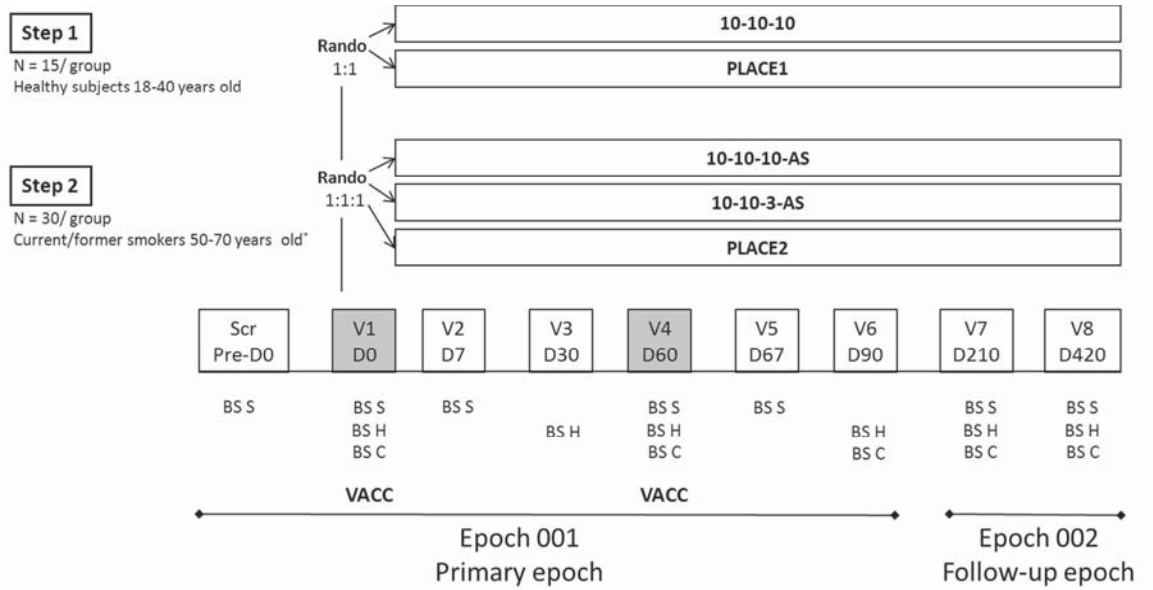
- To collect blood samples for assay development/validation and/or for evaluation/characterisation of the humoral and cellular immune responses to components of either the NTHi-Mcat investigational vaccines and/or of other respiratory pathogens.

3. STUDY DESIGN OVERVIEW

3.1. Study design

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 5.5), are essential and required for study conduct.

Figure 1 Study design overview



BS S = blood sample for safety (haematology/ biochemistry);
BS H = blood sample for humoral immune responses/assay development from all subjects of each step;
BS C = blood sample for cell-mediated immunogenicity from 50% of subjects in each group of Step 2;
Rando = randomisation; **Scr** = Screening Visit; **V** = Visit; **D** = Day; **VACC** = vaccination (Visits 1 and 4, indicated in grey);
* With a cigarette smoking history ≥10 pack-years (calculation pack-years = average number of cigarettes smoked per day x number of years smoked)/20

- **Experimental design**: Phase I, observer-blind, randomised, placebo-controlled, multi-centre study with two steps.
- **Duration of the study**: For each subject, the study will last approximately 15 months, from screening to study end.
  - Epoch 001: Primary starting at Screening Visit and ending at, and including Visit 6 (Day 90).
  - Epoch 002: Follow-up starting at Visit 7 (Day 210) and ending at Visit 8 (Day 420).

- **Study groups:**
  - **10-10-10**: Approximately 15 subjects receiving two doses of the non-adjuvanted GSK Biologicals' NTHi-Mcat investigational vaccine containing 10 µg of PD, PE-PilA and UspA2 during Step 1 of the study.
  - **PLACE1**: Approximately 15 subjects receiving two doses of placebo (saline solution) during Step 1 of the study.
  - **10-10-10-AS:** Approximately 30 subjects receiving two doses of the $AS01_E$-adjuvanted GSK Biologicals' NTHi-Mcat investigational vaccine containing 10 µg of PD, PE-PilA and UspA2 during Step 2 of the study.
  - **10-10-3-AS**: Approximately 30 subjects receiving two doses of the $AS01_E$-adjuvanted GSK Biologicals' NTHi-Mcat investigational vaccine containing 10 µg of PD, 10µg of PE-PilA, and 3.3 µg of UspA2 during Step 2 of the study.
  - **PLACE2**: Approximately 30 subjects receiving two doses of placebo (saline solution) during Step 2 of the study.

Table 1 Study groups and epochs foreseen in the study

Study groups	Number of subjects	Age (Min/Max)	Epochs		Step
			Epoch 001	Epoch 002	
10-10-10	15	18 - 40 years	x	x	1
PLACE1	15	18 - 40 years	x	x	1
10-10-10-AS	30	50 - 70 years	x	x	2
10-10-3-AS	30	50 - 70 years	x	x	2
PLACE2	30	50 - 70 years	x	x	2

Table 2 Study groups and treatment foreseen in the study

Treatment name	Vaccine/ product name	Study Groups				
		10-10-10	10-10-10-AS	10-10-3-AS	PLACE1	PLACE2
10-10-10/Plain	NTHi-Mcat 10-10-10	X				
	NaCl					
10-10-10/$AS01_E$	NTHi-Mcat 10-10-10		X			
	AS01E					
10-10-3/$AS01_E$	NTHi-Mcat 30-30-10 *			X		
	AS01E					
Placebo	NaCl				X	X

* Vials containing 37.5 µg, 37.5 µg and 12.5 µg of freeze-dried PD, PE-PilA and UspA2, respectively, will be diluted in three volumes of AS01E (total of ~1875 µL). 0.5 mL will be administered.

- **Control:** Placebo control.
- **Vaccination schedules:** 0, 2 months (i.e. at Visit 1 [Day 0] and Visit 4 [Day 60]).
- **Treatment allocation:** Subjects will be allocated to a study group using a centralised randomisation system on internet (SBIR). SBIR will also be used for treatment allocation of the second dose. The randomisation algorithm will use a minimisation procedure accounting for centre at Step 1 and accounting for age (50-59 years or

60-70 years), for smoking status (current or former smokers), for centre and for forced expiratory volume in 1 second (FEV_1) / forced vital capacity (FVC) (≥ 0.7 or < 0.7) at Step 2.

- **Blinding:** Observer-blind.

Table 3 Blinding of study epochs

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

- **Sampling schedule**:
  - Blood samples for **safety** (haematology/ biochemistry) will be collected from all subjects at Screening Visit (pre-Day 0), at Visit 1 (Day 0), Visit 2 (Day 7), Visit 4 (Day 60), Visit 5 (Day 67), Visit 7 (Day 210) and at Visit 8 (Day 420).
  - Blood samples for **immunogenicity** will be collected from all subjects for humoral immunity at Visit 1 (Day 0), Visit 3 (Day 30), Visit 4 (Day 60), Visit 6 (Day 90), Visit 7 (Day 210) and at Visit 8 (Day 420), and from a sub-cohort of subjects for cell-mediated immunity (CMI) at Visit 1 (Day 0), Visit 4 (Day 60), Visit 6 (Day 90), Visit 7 (Day 210) and at Visit 8 (Day 420).
- **Type of study:** Self-contained.
- **Data collection:** Electronic Case Report Form (eCRF).
- **Safety monitoring during vaccination period:** Safety evaluations by the Safety Review Team (SRT) (blinded) and by an iSRC (unblinded) will be performed. Refer to Section 8.10 for detailed description of holding rules and safety monitoring.

3.2. Safety precautions

3.2.1. Details of the staggered design of the study

This study will be conducted following a staggered design with two steps:

- **Step 1:** Vaccination of ~30 subjects randomised into the groups 10-10-10 and PLACE1 (1:1).
- **Step 2:** Vaccination of ~90 subjects randomised into the groups 10-10-10-AS, 10-10-3-AS and PLACE2 (1:1:1).

Please refer to Section 8.10 for detailed information about safety evaluations in this study.

3.2.2. Vaccination

In Step 1, for each of the vaccine doses (Dose 1 and Dose 2), all subjects will preferably be vaccinated within 2 weeks.

Limited vaccination

For Step 1, vaccination will be limited to 6 subjects/ day during the first three days for each vaccine dose. These subjects are to be vaccinated sequentially, leaving sufficient time for close observation for a minimum of 60 minutes prior to proceeding to the vaccination of the next subject. As soon as three days have passed and at least 12 subjects have been vaccinated, the remaining subjects from Step 1 can be vaccinated without further limitation.

Vaccination without limitation

After vaccination day 3 in Step 1 (see limited vaccination), and for all subjects in Step 2, there won't be a limitation in the number of vaccinees per day nor in the timing between vaccinations of each subject. All subjects will be closely observed for a minimum of 60 minutes after vaccination (Figure 2).

4. STUDY COHORT

4.1. Number of subjects/centres

This will be a multi-centre study.

There will be five study groups, three of which will receive an investigational NTHi-Mcat vaccine formulation and two placebo groups. The target is to enrol approximately 30 eligible subjects 18–40 years old in Step 1 (~15/group) and approximately 90 eligible subjects 50–70 years old in Step 2 (~30/group).The total target number of subjects is approximately 120 (see also Table 1).

Please refer to Section 10.3 for a detailed description of the criteria used in the determination of sample size.

Approximately 50% of the subjects in each group of Step 2 will be part of a **sub-cohort for CMI analysis**. ***Subjects will be selected based on the enrolment order, i.e. the first 50% of the participants (approximately the first 15 subjects/group) will be enrolled in the sub-cohort for CMI***. An additional blood sample for CMI analysis will be taken from those subjects at each of the CMI blood sampling time points (i.e. Day 0, Day 60, Day 90, Day 210 and Day 420). **(Amended 16 March 2016)**

Table 4 Sub-cohorts for CMI

Sub-cohort name	Description	Estimated number of subjects in Step 2
CMI	An additional blood sample will be taken from about 50% of subjects in each group of Step 2 for CMI analysis	~15/ group ~45 in total

CMI: cell-mediated immunity

4.2. Inclusion criteria for enrolment

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

4.2.1. Healthy adults 18-40 years old (Step 1)

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

- Subjects 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).
- A male or female between, and including, 18 and 40 years of age at the time of the Screening Visit.
- Written informed consent obtained from the subject.
- Healthy subjects without acute or chronic, clinically significant pulmonary, cardiovascular, hepatic or renal functional abnormality, as determined by physical examination or laboratory screening tests.
- Female subjects of non-childbearing potential may be enrolled in the study.
  - Non-childbearing potential is defined as pre-menarche, hysterectomy, ovariectomy or post-menopause.

  Please refer to the glossary of terms for the definition of menarche and menopause.
- Female subjects of childbearing potential may be enrolled in the study, if the subject:
  - has practiced adequate contraception for 30 days prior to vaccination, and
  - has a negative pregnancy test on the day of vaccination, and
  - has agreed to continue adequate contraception during the entire treatment period and for 2 months after completion of the vaccination series.

  Please refer to the glossary of terms for the definition of adequate contraception.

4.2.2. Current and former smokers 50-70 years old (Step 2)

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

- Subjects 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)**.**
- A male or female between, and including, 50 and 70 years of age at the time of the Screening Visit.
- Written informed consent obtained from the subject.
- Subjects without medical history, clinical finding or laboratory finding which in the opinion of the investigator could pose a safety concern or interfere with the protocol.
- Current or former smoker with a cigarette smoking history ≥ 10 pack-years.

  Please refer to the glossary of terms for the definitions of pack-years and of current and former smoker.
- Female subjects of non-childbearing potential may be enrolled in the study.
  - Non-childbearing potential is defined as pre-menarche, hysterectomy, ovariectomy or post-menopause.

  Please refer to the glossary of terms for the definition of menarche and menopause.
- Female subjects of childbearing potential may be enrolled in the study, if the subject:
  - has practiced adequate contraception for 30 days prior to vaccination, and
  - has a negative pregnancy test on the day of vaccination, and
  - has agreed to continue adequate contraception during the entire treatment period and for 2 months after completion of the vaccination series.

  Please refer to the glossary of terms for the definition of adequate contraception.

4.3. Exclusion criteria for enrolment

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

4.3.1. Healthy adults 18-40 years old (Step 1)

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:

- Use of any investigational or non-registered product (drug or vaccine) other than the study vaccines within 30 days preceding the first dose of study vaccine, or planned use during the study period.

- 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 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, with the exception of any influenza vaccine which may be administered ≥ 15 days preceding or following any study vaccine dose.
- Previous vaccination with any vaccine containing NTHi and/or Mcat antigens.
- Chronic administration (defined as more than 14 days in total) of immunosuppressants or other immune-modifying drugs within 6 months prior to the first vaccine dose. For corticosteroids, this will mean prednisone ≥20 mg/day, or equivalent. Only topical steroids are allowed.
- Administration of long-acting immune-modifying drugs at any time during the study period (e.g. infliximab).
- Administration of immunoglobulins and/or any blood products within the 3 months preceding the first dose of study vaccine or planned administration during the study period.
- Any confirmed or suspected immunosuppressive or immunodeficient condition, based on medical history and physical examination (no laboratory testing required).
- History of or current autoimmune disease.
- Laboratory evidence of clinically significant haematological (complete blood cell count [RBC, WBC], WBC differential count [lymphocytes, neutrophils and eosinophils], platelets count or haemoglobin level) and biochemical (alanine aminotransferase [ALT], aspartate aminotransferase [AST] or creatinine) abnormalities as per the opinion of the investigator.
- Acute disease and/or fever at the time of enrolment.
  - Fever is defined as temperature ≥ 37.5°C for oral or axillary route. The preferred route for recording temperature in this study will be oral.
  - Subjects with a minor illness (such as mild diarrhoea, mild upper respiratory infection) without fever may, be enrolled at the discretion of the investigator.
- Current alcoholism and/or drug abuse.

  Please refer to the glossary of terms for the definition of alcoholism.
- History of or current condition preventing intramuscular injection as bleeding or coagulation disorder.
- Malignancies within previous 5 years (excluding non-melanoma skin cancer) or lymphoproliferative disorders.
- Pregnant or lactating female.
- Female planning to become pregnant or planning to discontinue contraceptive precautions.

- Any other condition that the investigator judges may interfere with study findings.

4.3.2. Current or former smokers 50-70 years old (Step 2)

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:

- Use of any investigational or non-registered product (drug or vaccine) other than the study vaccines within 30 days preceding the first dose of study vaccine, or planned use during the study period.
- 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 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, with the exception of any influenza or pneumococcal licensed vaccine which may be administered ≥ 15 days preceding or following any study vaccine dose.
- Previous vaccination with any vaccine containing NTHi and/or Mcat antigens.
- Chronic administration (defined as more than 14 days in total) of immunosuppressants or other immune-modifying drugs within 6 months prior to the first vaccine dose. For corticosteroids, this will mean prednisone ≥20 mg/day, or equivalent. Only topical steroids are allowed.
- Administration of long-acting immune-modifying drugs at any time during the study period (e.g. infliximab).
- Administration of immunoglobulins and/or any blood products within the 3 months preceding the first dose of study vaccine or planned administration during the study period.
- Any confirmed or suspected immunosuppressive or immunodeficient condition, based on medical history and physical examination (no laboratory testing required).
- History of or current autoimmune disease.
- Post-bronchodilator FEV_1 < 80% of predicted normal value.
- Diagnosed with a respiratory disorder (e.g. asthma, COPD, sarcoidosis, tuberculosis, bronchiectasis, lung fibrosis, pulmonary embolism, pneumothorax, or physician-confirmed lung cancer). Please note that subjects with mild pulmonary obstruction (i.e. FEV_1/ FVC ratio < 0.7 with FEV_1 ≥ 80% of normal predicted values [GOLD grade 1]) can be enrolled.
- Has contraindication for spirometry testing (such as recent eye surgery, recent thoracic or abdominal surgery procedures, unstable cardiovascular status, recent myocardial infection or pulmonary embolism).

- Laboratory evidence of clinically significant* haematological (complete blood cell count [RBC, WBC], WBC differential count [lymphocytes, neutrophils and eosinophils], platelets count or haemoglobin level) and biochemical (ALT, AST or creatinine) abnormalities.

  * As the subjects are adult or elderly, current or former smokers, various pre-existing laboratory abnormalities might be detected. The investigator should use his clinical judgement to decide which ones are clinically significant.

- Acute disease and/or fever at the time of enrolment.
  - Fever is defined as temperature ≥ 37.5°C for oral or axillary route. The preferred route for recording temperature will be oral.
  - Subjects with a minor illness (such as mild diarrhoea, mild upper respiratory infection) without fever may be enrolled at the discretion of the investigator.

- Current alcoholism and/or drug abuse.

  Please refer to the glossary of terms for the definition of alcoholism.

- Has significant disease (including significant neurological or psychological disorders), in the opinion of the investigator, likely to interfere with the study and/or likely to cause death within the study duration.
- History of or current condition preventing intramuscular injection as bleeding or coagulation disorder.
- Malignancies within previous 5 years (excluding non-melanoma skin cancer) or lymphoproliferative disorders.
- Pregnant or lactating female.
- Female planning to become pregnant or planning to discontinue contraceptive precautions.
- Any other condition that the investigator judges may interfere with study findings.

5. CONDUCT OF THE STUDY

5.1. 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.

GSK will obtain favourable 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 favourable opinion/approval of study protocol and any subsequent amendments.
- Subject 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 informed consent must be obtained from each subject prior to participation in the study.

GSK Biologicals will prepare a model 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 judgement, 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.

5.2. Subject identification and randomisation of treatment

5.2.1. 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 centre.

5.2.2. Randomisation of treatment

5.2.2.1. Randomisation of supplies

Two randomisation lists corresponding to each step of the study will be generated at GSK Biologicals using MATerial Excellence (MATEX), a SAS® (Statistical Analysis System) programme developed by GSK Biologicals and will be used to number the vaccines. A block randomisation scheme will be used.

5.2.2.2. Treatment allocation to the subject

The treatment numbers will be allocated by dose.

5.2.2.2.1. Study group and treatment number allocation

The target will be to enrol approximately 120 eligible subjects who will be randomly assigned per step:

- **Step 1:** two study groups in a (1:1) ratio (approximately 15 subjects per group).
- **Step 2:** three study groups in a (1:1:1) ratio (approximately 30 subjects per group).

Allocation of the subject to a study group at the investigator site will be performed using SBIR.

Subjects will be minimised for centre at Step 1. At Step 2, subjects will be minimised for age (50-59 years or 60-70 years), for smoking status (current or former smokers), for centre and for forced expiratory volume in 1 second (FEV_1) / forced vital capacity (FVC) (≥ 0.7 or < 0.7). Minimisation factors will have equal weight in the minimisation algorithm.

As several minimisation factors have been defined for the treatment allocation at Step 2, it was decided at the design phase not to minimise for the CMI group allocation. This wouldn't be feasible from an implementation point of view. **(Amended 16 March 2016)**

After obtaining the signed and dated ICF from the subject (at Screening Visit) and after having checked the eligibility of the subject, the dedicated study staff will access SBIR (at Visit 1). Upon providing the information for each minimisation factor and the subject identification number, the randomisation system will determine the study group and will provide the treatment number to be used for the first dose. This treatment number will be used by the dedicated staff member for administration of the vaccine.

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 Study Procedures Manual (SPM) for specific instructions.

5.2.2.2.2. Treatment number allocation for subsequent doses

For the second dose, the dedicated study staff 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.

5.3. 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, and immunogenicity) will

all be unaware of which vaccine was administered. To do so, vaccine preparation and administration will be done by authorised medical personnel who will not participate in any of the study clinical evaluation assays.

An analysis of Epoch 001 will be done on as cleaned as possible data up to Visit 6 (Day 90) (please refer to Section 10.8.1 for more information). At this point, the GSK statistician will be unblinded (i.e. will have access to the individual subject treatment assignment). The remaining GSK study personnel (excluding the iSRC members) will stay blinded (i.e. will not have access to the individual subject treatment assignment) until study end. It is possible however, due to the limited sample size, that unblinding occurs for a few subjects having a specific adverse event (AE) or serious AE (SAE) (e.g. an AE/SAE occurring only in a single group). Therefore anyone having access to the analysis of Epoch 001 could become unblinded regarding those specific cases.

The site staff will work in an observer-blind manner during the entire study. As the vaccines in this study are of different appearance (see also Table 14), two teams of study personnel will be set up:

- A team of unblinded personnel (responsible for the reception, preparation and administration of the vaccines).
- A team of blinded personnel (responsible for the clinical safety evaluation of the subjects).

Please refer to the SPM for more information on vaccine preparation and administration while maintaining the blind.

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.

The laboratory personnel in charge of CMI testing may be unblinded for treatment (active treatment versus placebo) due to the observed response in the CMI samples of certain subjects.

5.4. 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.

Refer to Section 8.10 for more information on safety holding rules and safety evaluation by iSRC.

5.5. Outline of study procedures

Table 5 List of study procedures for healthy adults 18-40 years old (Step 1)

Epoch	*Epoch 001*							*Epoch 002*	
Type of contact	**Screening Visit**	**Visit 1**	**Visit 2**	**Visit 3**	**Visit 4**	**Visit 5**	**Visit 6**	**Visit 7**	**Visit 8**
Time points	**pre-Day 0**	**Day 0**	**Day 7**	**Day 30**	**Day 60**	**Day 67**	**Day 90**	**Day 210**	**Day 420**
Sampling time points	**Screening**	**Pre**	**Post-Vacc I**			**Post-Vacc II**			
Informed consent	●								
Check inclusion/exclusion criteria	●(d)	O							
Collect demographic data	●								
Medical history	O(d)	●							
Measure/record height and weight	●								●
Physical examination	●(d, e)	O	O	O	O	O	O	O	O
Urine pregnancy test (a)	●	●			●				
Check contraindications		O			O				
Pre-vaccination body temperature		●			●				
Treatment number allocation (b)		●			●				
Blood sampling for safety assessment (~5.5 mL)	●(d)	●	●		●	●		●	●
Blood sampling for antibody determination and assay development (~20 mL)		●		●	●		●	●	●
Vaccine administration		●			●				
Distribution of Diary Cards (c)		O	O		O	O			
Return of Diary Cards			O	O		O	O		
Diary Card transcription by investigator			●	●		●	●		
Recording of AEs		●	●	●	●	●	●		
Recording of SAEs	●(f)	●	●	●	●	●	●	●	●
Recording of pIMDs		●	●	●	●	●	●	●	●
Recording of pregnancies		●	●	●	●	●	●	●	●
Record concomitant medication/vaccination (g)		●	●	●	●	●	●	●	●
Record intercurrent medical conditions requiring medical attention		●	●	●	●	●	●	●	●
Screening conclusion	●								
Study conclusion									●

Note: The double-line borders following Day 90 and Day 420 indicates the statistical analyses which will be performed on all data (i.e. data that are as clean as possible) obtained up to Day 90 and Day 420, respectively.
Vacc: Vaccination; **AEs:** Adverse events; **SAEs:** Serious adverse events; **pIMDs:** potential immune-mediated disease.
● 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.
(a) A urine pregnancy test will be performed only for women of childbearing potential.
(b) Treatment number allocation with randomisation at Visit 1 (Day 0); treatment number allocation without randomisation at Visit 4 (Day 60).
(c) Distribution of **2** Diary Cards after each vaccination:

- a 1st one distributed on the day of vaccination for recording of solicited and unsolicited AEs and concomitant medications/ products and vaccinations from Day 0 to Day 6 after each vaccination.
- a 2nd one distributed on the 7 day post-vaccination visit for recording of unsolicited AEs and concomitant medications/ products and vaccinations from Day 7 to Day 29 after each vaccination.

(d) Screening evaluations may be completed 1 to 28 days before Day 0. Site staff should allow sufficient time between the Screening and Visit 1 to receive and review screening safety laboratory test results. If a delay occurs such that the interval between Screening and the Visit 1 vaccination exceeds 28 days, a re-screening visit should be scheduled before Visit 1 during which blood sample collection for safety laboratory assessment must be repeated; an interim medical history and physical examination must be obtained and inclusion / exclusion criteria must be re-reviewed.
(e) Complete physical examination including vital signs.

(f) From Screening to Visit 1, only those SAEs that are considered related to study participation or to concomitant use of GSK products or any fatal events need to be recorded. GSK products or any fatal event need to be recorded.
(g) Concomitant medication products/vaccination as indicated in Section 6.6.1 need to be recorded.

Table 6 List of study procedures for current or former smokers 50-70 years old (Step 2) (Amended 16 March 2016)

Epoch	*Epoch 001*							*Epoch 002*	
Visit 6	**Screening Visit**	**Visit 1**	**Visit 2**	**Visit 3**	**Visit 4**	**Visit 5**	**Visit 6**	**Visit 7**	**Visit 8**
Time points	**pre-Day 0**	**Day 0**	**Day 7**	**Day 30**	**Day 60**	**Day 67**	**Day 90**	**Day 210**	**Day 420**
Sampling time points	**Screening**	**Pre**	**Post-Vacc I**			**Post-Vacc II**			
Informed consent	●								
Check inclusion/exclusion criteria	●(e)	O							
Collect demographic data	●								
Medical history	O(e)	●							
Smoking status	●							●	●
Smoking exposure history	●								
Measure/record height and weight	●								●
Spirometry *(i)*	●								●
Physical examination	●(e, f)	O	O	O	O	O	O	O	O
Urine pregnancy test (a)	●	●			●				
Check contraindications		O			O				
Pre-vaccination body temperature		●			●				
Treatment number allocation(b)		●			●				
Blood sampling for safety assessment (~5.5 mL)	●(e)	●	●		●	●		●	●
Blood sampling for antibody determination and assay development (~20 mL)		●		●	●		●	●	●
Blood sampling for CMI response (~20 mL)(c)		●			●		●	●	●
Vaccine administration		●			●				
Distribution of Diary Cards(d)		O	O		O	O			
Return of Diary Cards			O	O		O	O		
Diary Card transcription by investigator			●	●		●	●		
Recording of AEs		●	●	●	●	●	●		
Recording of SAEs	●(g)	●	●	●	●	●	●	●	●
Recording of pIMDs		●	●	●	●	●	●	●	●
Recording of pregnancies		●	●	●	●	●	●	●	●
Record concomitant medication/vaccination(h)		●	●	●	●	●	●	●	●
Record intercurrent medical conditions requiring medical attention		●	●	●	●	●	●	●	●
Screening conclusion	●								
Study conclusion									●

Note: The double-line borders following Day 90 and Day 420 indicates the statistical analyses which will be performed on all data (i.e. data that are as clean as possible) obtained up to Day 90 and Day 420, respectively.
Vacc: Vaccination; **CMI:** Cell-mediated immunity; **AEs:** Adverse events; **SAEs:** Serious adverse events; **pIMDs:** potential immune-mediated disease.
● 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.
(a) A urine pregnancy test will be performed only for women of childbearing potential.
(b) Treatment number allocation with randomisation at Visit 1 (Day 0); treatment number allocation without randomisation at Visit 4 (Day 60).
(c) Only for subjects belonging to the CMI sub-cohort.
(d) Distribution of **2** Diary Cards after each vaccination:

- a 1st one distributed on the day of vaccination for recording of solicited and unsolicited AEs and concomitant medications/ products and vaccinations from Day 0 to Day 6 after each vaccination.
- a 2nd one distributed on the 7 day post-vaccination visit for recording of unsolicited AEs and concomitant medications/ products and vaccinations from Day 7 to Day 29 after each vaccination.

(e) Screening evaluations may be completed 1 to 28 days before Day 0. Site staff should allow sufficient time between the Screening and Visit 1 to receive and review screening safety laboratory test results. If a delay occurs such that the interval between Screening and the Visit 1 vaccination exceeds 28 days, a re-screening visit should be scheduled before Visit 1 during which blood sample collection for safety laboratory assessment must be repeated; an interim medical history and physical examination must be obtained and inclusion / exclusion criteria must be re-reviewed.
(f) Complete physical examination including vital signs.
(g) From Screening to Visit 1, only those SAEs that are considered related to study participation or to concomitant use of GSK products or any fatal events need to be recorded. GSK products or any fatal event need to be recorded.
(h) Concomitant medication products/vaccination as indicated in Section 6.6.1 need to be recorded.
(i) ***This test should be repeated on another day if the first attempt doesn't meet quality requirements as described by ATS/ERS standards (Miller, 2005).***

Table 7 Intervals between study visits

Interval			Optimal length of interval (a)	Allowed interval
Screening (Pre-Day 0)	→	Visit 1 (Day 0)	5 days	1 – 28 days (b)
Visit 1 (Day 0)	→	Visit 2 (Day 7)	7 days	7 - 9 days
Visit 1 (Day 0)	→	Visit 3 (Day 30)	30 days	30 - 40 days
Visit 1 (Day 0)	→	Visit 4 (Day 60)	60 days	60 - 70 days
Visit 4 (Day 60)	→	Visit 5 (Day 67)	7 days	7 - 9 days
Visit 4 (Day 60)	→	Visit 6 (Day 90)	30 days	30 - 40 days
Visit 1 (Day 0)	→	Visit 7 (Day 210)	210 days	210 - 230 days
Visit 1 (Day 0)	→	Visit 8 (Day 420)	420 days	420 - 440 days

Note: visits out of range can lead to elimination from ATP analyses.
(a) Whenever possible the investigator should arrange study visits within this interval.
(b) Screening evaluations may be completed 1 to 28 days before Day 0. Site staff should allow sufficient time between the Screening and Day 0 visits to receive and review screening safety laboratory test results. If a delay occurs such that the interval between Screening and the Day 0 vaccination exceeds 28 days, a re-screening visit should be scheduled before Visit 1 during which blood sample collection for safety laboratory assessment must be repeated; an interim medical history and physical examination must be obtained and inclusion / exclusion criteria must be re-reviewed.

5.6. Detailed description of study procedures

5.6.1. Informed consent

The signed informed consent of the subject must be obtained before study participation. Refer to Section 5.1 for the requirements on how to obtain informed consent.

5.6.2. Check inclusion and exclusion criteria

Check all inclusion and exclusion criteria as described in Sections 4.2 and 4.3 before enrolment.

5.6.3. Collect demographic data

Record demographic data such as year of birth, gender and race (ethnicity and geographic ancestry) in the subject's eCRF.

5.6.4. 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.

5.6.5. Smoking status

For all subjects in Step 2, record smoking status (current or former smoker) in the subject's eCRF.

Please refer to the glossary of terms for the definitions of current and former smoker.

5.6.6. Smoking exposure history

For all subjects in Step 2, smoking history will be obtained by means of a self-administered questionnaire (which will be part of the ATS-DLD-78A questionnaire) provided to the subjects, in which they will give answers about their smoking history, including duration (number of years) and number of cigarettes smoked.

Please refer to the SPM for more details on questionnaire.

From the information obtained via the questionnaire, calculate the pack-years using the following calculation (also refer to the glossary of terms for the definitions of pack-years) Please note that pipe and/or cigar use should not be used to calculate pack-year history:

$$\text{Total pack-years} = \frac{\text{Average no. cigarettes smoked / day x no. years of smoking}}{20}$$

All data will be recorded in the subject's eCRF.

5.6.7. Measure/record height and weight

Measure height and weight of the subject and record the data in the 'physical examination' section of the eCRF.

5.6.8. Spirometry

For all subjects in Step 2, spirometry to assess FEV_1 and FVC will be performed using techniques that meet American Thoracic Society (ATS)/European Respiratory Society (ERS) published standards [Miller, 2005].

Only certified study staff can perform spirometry assessment.

The spirometry test must be of good quality. If this is not the case, a re-test should be done. **(Amended 16 March 2016)**

The following measurements (best efforts)/ values will be used:

- FVC, which is the total volume of air that a person exhales in a forced expiratory manoeuvre (the act of exhaling as hard and fast as possible after maximal inspiration), measured in litres.
- FEV_1, which is the amount of air that a person breathes out during the first second of a forced expiratory manoeuvre, measured in litres.
- FVC predicted normal value.
- FEV_1 predicted normal value.
- FEV_1 % of predicted normal value.
- The ratio of FEV_1 to the FVC (FEV_1/FVC).

If $FEV_1 < 80\%$ of predicted normal values and/ or FEV_1/ FVC ratio < 0.7 without bronchodilator, post-bronchodilator spirometry should also be performed.

The data will be directly transferred from the provider to GSK Biologicals.

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

5.6.9. Physical examination

Perform a complete physical examination of the subject at the Screening Visit, including resting vital signs (systolic/diastolic blood pressure, heart rate and respiratory rate after at least 10 minutes of rest). Collected information needs to be recorded in the eCRF.

Physical examination at each study visit subsequent to the Screening Visit will be performed only if the subject indicates that there might be some underlying pathology(ies) or if deemed necessary by the investigator or delegate.

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.

5.6.10. Urine pregnancy test

Female subjects of childbearing potential are to have a urine pregnancy test prior to any study vaccine administration. The study vaccines may only be administered if the pregnancy test is negative.

Note: The urine pregnancy test must be performed even if the subject is menstruating at the time of the study visit.

5.6.11. Check contraindications

Contraindications, warnings and precautions to vaccination must be checked at the beginning of each vaccination visit. Refer to Sections 6.5 for more details.

5.6.12. Assess pre-vaccination body temperature

The oral or axillary body temperature of all subjects needs to be measured prior to any study vaccine administration. The preferred route for recording temperature in this study will be oral.

If the subject has fever (fever is defined as a temperature ≥ 37.5°C for oral or axillary route) on the day of vaccination, the vaccination visit will be rescheduled within the allowed interval for this visit (see Table 7).

5.6.13. Study group and treatment number allocation

Study group and treatment number allocation will be performed as described in Section 5.2.2. The number of each administered treatment must be recorded in the eCRF.

5.6.14. Blood sampling

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

5.6.14.1. Blood sampling for safety assessment

Blood samples for safety assessment will be taken from all subjects during certain study visits as specified in Table 5 and Table 6 ‘List of study procedures’.

A volume of approximately 5.5 mL of whole blood should be drawn at each pre-defined time point.

When borderline abnormalities in haematology or biochemistry parameters (i.e. considered not clinically significant) are reported at Screening Visit, at least one re-test (at least one week after the first result) has to be done before Visit 1 to ensure that the abnormality is not due to an evolving medical condition.

Blood samples for safety assessment may be repeated during the course of the study at discretion of the investigator if medically indicated.

5.6.14.2. Blood sampling for antibody determination and assay development

Blood samples for antibody determination and assay development will be taken from all subjects during specific study visits as specified in Table 5 and Table 6.

A volume of approximately 20 mL of whole blood would be drawn at each pre-defined time point. After centrifugation, the serum should be stored at -20°C or colder until shipment. Refer to the SPM and to the central laboratory manual for more details on sample storage conditions.

5.6.14.3. Blood sampling for CMI

Blood samples for CMI will be taken from the subjects in the sub-cohort for CMI analysis, and for development of read-outs during certain study visits as specified in Table 6.

A volume of approximately 20 mL of whole blood should be drawn from all subjects included in the immunogenicity sub-cohort for CMI at each pre-defined time point. The blood should be stored at the investigator's site at room temperature and it must not be centrifuged. Samples will be shipped at room temperature (20 to 25°C) to the designated laboratory for cell and plasma separation to be performed within 24 hours of collection.

5.6.15. Vaccination

After completing all prerequisite procedures prior to vaccination, one dose of study vaccine will be administered intramuscularly (IM) in the deltoid of the non-dominant arm (refer to Section 6.3 for detailed description of the vaccine administration procedure).

If the investigator or delegate determines that the subject'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 5 and Table 6).

The subjects will be observed closely for at least 60 minutes following the administration of the vaccine, with appropriate medical treatment readily available in case of anaphylaxis.

5.6.16. Recording of AEs, SAEs, pregnancies and pIMDs

Refer to Section 8.3 for procedures for the investigator to record AEs, SAEs, pregnancies and pIMDs. Refer to Section 8.4 for guidelines on how to submit SAE, pregnancy and pIMD reports to GSK Biologicals.

The subjects will be instructed to contact the investigator immediately should they manifest any signs or symptoms they perceive as serious.

Diary Cards

- After each vaccination, two Diary Cards will be provided to the subjects:

  - The **first** Diary Card will be provided at the vaccination visit. On this Diary Card, the subject will record any solicited local/general AEs, any unsolicited AEs and all concomitant medications/ products and vaccinations during a 7-day follow-up period (i.e. on the day of vaccination and during the next 6 days). The subject will be instructed to return the completed diary card to the investigator at the next study visit (7 days post-vaccination).
  - The **second** Diary Card will be provided at the next study visit (7 days post-vaccination). On this Diary Card, the subject will record any unsolicited AEs and all concomitant medications/ products and vaccinations from Day 7 to Day 29 after each vaccination (for 30-day follow-up period). The subject will be instructed to return the completed diary card to the investigator at the next study visit (30 days post-vaccination).

- Collect and verify completed Diary Cards during discussion with the subject during each 7-days post-vaccination visit and during each 30-day post-vaccination visit.
- Any unreturned Diary Cards will be sought from the subject through telephone call(s) or any other convenient procedure.
- The investigator will transcribe the collected information into the eCRF in English.

Refer to the SPM for more details on Diary Card completion guidelines.

5.6.17. Check and record concomitant medication/vaccination and intercurrent medical conditions requiring medical attention

Concomitant medication/vaccination must be checked and recorded in the eCRF as described in Section 6.6.

Intercurrent medical conditions must be checked and recorded in the eCRF as described in Section 6.7.

5.6.18. Study conclusion

The investigator will:

- review data collected to ensure accuracy and completeness.
- complete study conclusion screen in the eCRF.

5.7. Biological sample handling and analysis

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

Samples will not be labelled 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.

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).

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.

5.7.1. Use of specified study materials

When materials are provided by GSK Biologicals ***or by the Central laboratory***, 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 10.4 for the definition of cohorts to be analysed). 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. **(Amended 16 March 2016)**

5.7.2. Biological samples

Table 8 Biological samples

Sample type	Quantity	Unit	Time point	Sub-cohort
Blood for safety assessment	~5.5	mL	Screening Visit (pre-Day 0)	All screened subjects
			Visit 1 (Day 0)	All enrolled subjects
			Visit 2 (Day 7)	
			Visit 4 (Day 60)	
			Visit 5 (Day 67)	
			Visit 7 (Day 210)	
			Visit 8 (Day 420)	
Blood for antibody determination and assay development	~20	mL	Visit 1 (Day 0))	All enrolled subjects
			Visit 3 (Day 30)	
			Visit 4 (Day 60)	
			Visit 6 (Day 90)	
			Visit 7 (Day 210)	
			Visit 8 (Day 420)	
Blood for CMI response	~20	mL	Visit 1 (Day 0))	Sub-cohort for CMI*
			Visit 4 (Day 60)	
			Visit 6 (Day 90)	
			Visit 7 (Day 210)	
			Visit 8 (Day 420)	

* Refer to Section 4.1 for sub-cohort description.

5.7.3. 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 used for sample analysis.

Haematology and biochemistry

Haematology and biochemistry assays for safety assessment will be performed in a central laboratory (refer to Table 9).

Table 9 Haematology and biochemistry tests

System	Discipline	Component	Method	Scale	Laboratory**
Whole blood	Haematology	Leukocytes (White Blood Cells)	Per contract laboratory's procedures	Quantitative	Central laboratory
		Neutrophils*	Per contract laboratory's procedures	Quantitative	
		Lymphocytes*	Per contract laboratory's procedures	Quantitative	
		Basophils*	Per contract laboratory's procedures	Quantitative	
		Monocytes*	Per contract laboratory's procedures	Quantitative	
		Eosinophils*	Per contract laboratory's procedures	Quantitative	
		Haemoglobin	Per contract laboratory's procedures	Quantitative	
		Platelets	Per contract laboratory's procedures	Quantitative	
		Erythrocytes (Red Blood Cells)	Per contract laboratory's procedures	Quantitative	
Serum	Biochemistry	Alanine Aminotransferase (ALT)	Per contract laboratory's procedures	Quantitative	Central laboratory
		Aspartate Aminotransferase (AST)	Per contract laboratory's procedures	Quantitative	
		Creatinine	Per contract laboratory's procedures	Quantitative	

* For White Blood Cell (WBC) differential count.
** Refer to Table 25 for the laboratory address.

Humoral immune responses

Serological assays for the quantification of antibodies will be performed by ELISA at GSK Biologicals' laboratory or in a laboratory designated by GSK Biologicals using standardised procedures (refer to Table 10).

Table 10 Humoral Immunity (antibody determination) (Amended 16 March 2016)

System	Component	Method	Kit / Manufacturer	Unit	Cut-off	Laboratory*
Serum	Anti-PD	ELISA	In house	EL.U/mL	153	GSK Biologicals**
Serum	Anti-PE	ELISA	In house	EL.U/mL	8	GSK Biologicals**
Serum	Anti-PilA	ELISA	In house	EL.U/mL	7	GSK Biologicals**
Serum	Anti-UspA2	ELISA	In house	EL.U/mL	18	GSK Biologicals**

ELISA: Enzyme-Linked Immunosorbent Assay; EL.U/mL = ELISA unit per millilitre;
* Refer to APPENDIX B for the laboratory addresses.
** GSK Biologicals laboratory refers to the ***Clinical Laboratory Sciences (CLS) Laboratories*** in Rixensart, Belgium; Wavre, Belgium or a designated laboratory.

Serum samples might also be used for **development and validation of other assays** to measure the immune response to components of either the NTHi-Mcat investigational vaccines and/or of other respiratory pathogens such as, but not limited to, serum bactericidal activity assay, anti-PD, -PE, -PilA, -UspA2 functional assays, immunochemistry assays for other immunoglobulin classes and/or for anti-IgG subclasses, and anti-$AS01_E$ assays. Hence, additional use and/or testing of serum samples may occur during the course of the study or after study completion in case these data should be required for accurate interpretation of the study results and/or for further research related to the vaccine and/ or the disease and/ or should such test(s) become available at GSK Biologicals' laboratory or a laboratory designated by GSK Biologicals.

Cell-mediated immune responses

CMI assays will be performed at GSK Biologicals' laboratory using standardised procedures (refer to Table 11). Plasma collected during the processing of peripheral blood mononuclear cells (PBMCs) will be stored for potential future analyses and/or for assay development purposes.

Table 11 Cell-Mediated Immunity (CMI) (Amended 16 March 2016)

System	Component	Scale	Method	Unit	Laboratory*
PBMC	Specific $CD4^+/CD8^+$ T-cells	Quantitative	Flow cytometry	Number of specific $CD4^+/CD8^+$ T cells /10^6	GSK Biologicals**

PBMC = peripheral blood mononuclear cell.
* Refer to APPENDIX B for the laboratory addresses.
** GSK Biologicals laboratory refers to the ***Clinical Laboratory Sciences (CLS) Laboratories*** in Rixensart, Belgium; Wavre, Belgium or a designated laboratory.

Additional testing on PBMCs, such as, but not limited to, evaluation of NTHi- and Mcat-specific memory B-cells and intracellular cytokine staining (ICS) using other microbial antigens, may be done during the study or after study completion, should these data be required for accurate interpretation of the data and/ or for further research related to the investigational vaccine and/ or the disease and/ or should such test(s) become available at GSK Biologicals' laboratory or a laboratory designated by GSK Biologicals.

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.

5.7.4. Biological samples evaluation

5.7.4.1. Immunological read-outs

Table 12 Immunological read-outs

Blood sampling time point		Sub-cohort Name	No. subjects	Component
Type of contact and time point	Sampling time point			
Visit 1 (Day 0)	Pre-Vacc	All subjects	~120	Anti-PD, Anti-PE, Anti-PilA and Anti-UspA2
		CMI sub-cohort*	~45	Specific $CD4^+/CD8^+$ T-cells
Visit 3 (Day 30)	Post-Vacc I	All subjects	~120	Anti-PD, Anti-PE, Anti-PilA and Anti-UspA2
Visit 4 (Day 60)	Post-Vacc I	All subjects	~120	Anti-PD, Anti-PE, Anti-PilA and Anti-UspA2
		CMI sub-cohort*	~45	Specific $CD4^+/CD8^+$ T-cells
Visit 6 (Day 90)	Post-Vacc II	All subjects	~120	Anti-PD, Anti-PE, Anti-PilA and Anti-UspA2
		CMI sub-cohort*	~45	Specific $CD4^+/CD8^+$ T-cells
Visit 7 (Day 210)	Post-Vacc II	All subjects	~120	Anti-PD, Anti-PE, Anti-PilA and Anti-UspA2
		CMI sub-cohort*	~45	Specific $CD4^+/CD8^+$ T-cells
Visit 8 (Day 420)	Post-Vacc II	All subjects	~120	Anti-PD, Anti-PE, Anti-PilA and Anti-UspA2
		CMI sub-cohort*	~45	Specific $CD4^+/CD8^+$ T-cells

* Approximately 50% of the subjects in each group of the Step 2 will be part of a sub-cohort for CMI analysis.

5.7.4.2. Haematology and biochemistry

Table 13 Haematology/ biochemistry

Blood sampling time point		Sub-cohort Name	No. subjects	Component
Type of contact and time point	Sampling time point			
Screening* (Pre-Day 0)	Screening	All subjects	~120	**Haematology** (leukocytes [white blood cells], white blood cell differential count [neutrophils, lymphocytes, eosinophils, basophils, monocytes], erythrocytes [red blood cells], haemoglobin, platelets **Biochemistry** (ALT, AST, creatinine)
Visit 1 (Day 0)	Pre-Vacc I	All subjects	~120	
Visit 2 (Day 7)	Post-Vacc I	All subjects	~120	
Visit 4 (Day 60)	Post-Vacc I	All subjects	~120	
Visit 5 (Day 67)	Post-Vacc II	All subjects	~120	
Visit 7 (Day 210)	Post-Vacc II	All subjects	~120	
Visit 8 (Day 420)	Post-Vacc II	All subjects	~120	

ALT: Alanine aminotransferase; AST: Aspartate aminotransferase
Screening evaluations may be completed 1 to 28 days before Day 0. Site staff should allow sufficient time between the Screening and Day 0 visits to receive and review screening safety laboratory test results. If a delay occurs such that the interval between Screening and the Day 0 vaccination exceeds 28 days, a re-screening visit should be scheduled before Visit 1 during which blood sample collection for safety laboratory assessment should be repeated.

5.7.5. Immunological correlates of protection

No generally accepted immunological correlate of protection has been demonstrated so far for the antigens used in the investigational vaccines.

6. STUDY VACCINES AND ADMINISTRATION

6.1. Description of study vaccines

All candidate vaccines to be used have been developed and manufactured by GSK Biologicals.

The Quality Control Standards and Requirements for each 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 labelled and packed according to applicable regulatory requirements.

Table 14 Study vaccines

Treatment name	Vaccine/ product name	Formulation	Presentation	Volume to be administered	No doses
10-10-10/ Plain	NTHi-Mcat 10-10-10	PD=10 µg; PE-PilA=10 µg; UspA2=10 µg	Freeze-dried antigens in monodose vial	0.5 ml	2
	NaCl	NaCl=150mM	Liquid in monodose vial		
10-10-10/AS01$_E$	NTHi-Mcat 10-10-10	PD=10 µg; PE-PilA=10 µg; UspA2=10 µg	Freeze-dried antigens in monodose vial	0.5 ml	2
	AS01E	MPL=25 µg; QS21=25 µg; Liposomes	Liquid in monodose vial		
10-10-3/AS01$_E$	NTHi-Mcat 30-30-10	* PD=30 µg; PE-PilA=30 µg; UspA2=10 µg	Freeze-dried antigens in monodose vial	0.5 ml	2
	AS01E	MPL=25 µg; QS21=25 µg; Liposomes	Liquid in monodose vial		
Placebo	NaCl	NaCl=150mM	Liquid in monodose vial	0.5 ml	2

*Vials containing 37.5 µg, 37.5 µg and 12.5 µg of freeze-dried PD, PE-PilA and UspA2, respectively, will be diluted in three volumes of AS01$_E$ (total of ~1875 µL). 0.5 mL will be administered.

6.2. Storage and handling of study vaccines

The study vaccines 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.

Any temperature excursion outside the range of 0.0 to +8.0°C (for +2 to +8°C label storage condition) impacting investigational medicinal products (IMPs) 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.

In case of temperature excursion below +2.0°C down to 0.0°C impacting IMP(s) there is no need to report in (e)TDF, but adequate actions must be taken to restore the +2 to +8°C label storage temperature conditions. The impacted IMP(s) may still be administered, but the site should avoid re-occurrence of such temperature excursion. Refer to the Module on Clinical Trial Supplies in the SPM for more details on actions to take.

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.

6.3. Dosage and administration of study vaccines

Table 15 Dosage and administration

Type of contact and time point	Volume to be administered	Study Group	Treatment name	Route [1]	Site	Side [2]
Visit 1 (Day 0)	0.5mL	10-10-10	10-10-10/Plain	IM	Deltoid	Non-dominant
		10-10-10-AS	10-10-10/AS01$_E$			
		10-10-3-AS	10-10-3/AS01$_E$*			
		PLACE1	Placebo			
		PLACE2				
Visit 4 (Day 60)	0.5mL	10-10-10	10-10-10/Plain	IM	Deltoid	Non-dominant
		10-10-10-AS	10-10-10/AS01$_E$			
		10-10-3-AS	10-10-3/AS01$_E$*			
		PLACE1	Placebo			
		PLACE2				

[1] Intramuscular (IM)

[2] The deltoid of the non-dominant arm is the preferred side of injection. In case it is not possible to inject in the non-dominant arm, an injection in the dominant arm may be performed.

* Vials containing 37.5 μg, 37.5 μg and 12.5 μg of freeze-dried PD, PE-PilA and UspA2, respectively, will be diluted in three volumes of AS01$_E$ (~1875 μL). 0.5mL will be administered.

6.3.1. Instructions for study vaccine preparation and administration

GSK Biologicals' NTHi-Mcat investigational vaccines should be prepared maximum two hours prior to injection.

6.3.1.1. Instructions for reconstitution and administration of investigational 10-10-10/Plain or 10-10-10/AS01$_E$ vaccines

The vaccine formulations 10-10-10/Plain and 10-10-10/AS01$_E$ will be delivered as two separate monodose vials, one containing the freeze-dried cake (NTHi-Mcat 10-10-10) and the other one containing the diluent (saline or AS01$_E$ adjuvant). The entire content of the liquid-containing vial (diluent) will be injected into the vial of the freeze-dried cake. Following complete reconstitution of the cake by gentle rotations, the entire volume of this final reconstituted formulation will be withdrawn into a syringe. The excess volume

together with any remaining air bubbles will be expelled, so that a volume of 0.5 mL is finally administered to the subject. The content of the syringe will be masked with an opaque injection label to ensure blinding of the subjects. A small gap at the top of the syringe should be left in order to allow the administrator of the vaccine to check the liquid before injection. Finally, intramuscular injection of 0.5 mL should be done into the deltoid of the non-dominant arm. In case it is not possible to inject in the non-dominant arm, an injection in the dominant arm may be performed.

6.3.1.2. Instructions for reconstitution and administration of investigational 10-10-3/AS01$_E$ vaccine

The vaccine formulation 10-10-3/AS01$_E$ will be delivered as four separate monodose vials, one containing the freeze-dried cake (NTHi-Mcat 30-30-10) and three vials containing the AS01$_E$ adjuvant. The whole content of each of the three vials of AS01$_E$ must be used to reconstitute one vial of NTHi-Mcat 30-30-10 freeze-dried cake. Complete reconstitution of the cake by gentle rotations needs to be achieved before vaccine can be retrieved for injection. A suitable volume of this final reconstituted formulation will be withdrawn in a new calibrated syringe. The excess volume together with any remaining air bubbles will be expelled, so that a volume of 0.5 mL is administered to the subject. The content of the syringe should be masked with an opaque injection label to ensure blinding of the subjects. A small gap at the top of the syringe should be left in order to allow the administrator of the vaccine to check the liquid before injection. Finally, intramuscular injection of 0.5 mL should be done into the deltoid of the non-dominant arm. In case it is not possible to inject in the non-dominant arm, an injection in the dominant arm may be performed.

Please refer to the SPM for more detailed instructions on study vaccines preparation (e.g. needles and syringes used for reconstitution).

6.3.1.3. Instructions for preparation and administration of saline placebo

The saline solution will be delivered in a monodose vial, ready to use. The saline solution is transparent. If turbid or a precipitation is observed, the vial should not be used. Air bubbles and any excess volume have to be expelled. The content of the syringe will be masked with an opaque injection label to ensure blinding of the subjects. A small gap at the top of the syringe should be left in order to allow the administer to check the liquid before injection. Finally, intramuscular injection of 0.5 mL should be done into the deltoid of the non-dominant arm. In case it is not possible to inject in the non-dominant arm, an injection in the dominant arm may be performed.

Please refer to the SPM for more detailed instructions on study vaccines administration.

6.4. Replacement of unusable vaccine doses

In addition to the vaccine doses provided for the planned number of subjects (including over-randomisation when applicable), at least 20% 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 randomisation.

6.5. Contraindications to subsequent vaccination

The following events constitute absolute contraindications to further administration of the NTHi-Mcat investigational vaccines. If any of these events occur during the study, the subject must not receive vaccine Dose 2 but may continue other study procedures at the discretion of the investigator (see Section 8.5).

- Anaphylaxis following the administration of vaccine.
- Pregnancy.
- Any confirmed or suspected immunosuppressive or immunodeficient condition, including HIV infection.
- Current auto-immune disease.

The following events constitute contraindications to administration of the NTHi-Mcat investigational vaccines 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 Table 7), or the subject may be withdrawn at the discretion of the investigator (see Section 8.5).

- Acute disease and/or fever at the time of vaccination.
  - Fever is defined as a temperature ≥ 37.5°C for oral or axillary route. The preferred route for recording temperature in this study will be oral.
  - Subjects with a minor illness (such as mild diarrhoea, mild upper respiratory infection) without fever can be administered all vaccines.

6.6. Concomitant medications/products and concomitant vaccinations

At each study visit/contact, the investigator should question the subject about any medication/product taken and vaccination received by the subject.

6.6.1. Recording of concomitant medications/products and concomitant vaccinations

The following concomitant medications/products/vaccines must be recorded in the eCRF if administered during the indicated recording period:

- All concomitant medications/products, except vitamins and dietary supplements, administered 30 days following each dose of study vaccine.
- Any concomitant vaccination administered in the period starting 30 days before the first dose of study vaccine and ending at the last study visit.
- Prophylactic medication (i.e. medication administered in the absence of ANY symptom and in anticipation of a reaction to the vaccination).

  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 an oral or axillary temperature ≥ 37.5°C for oral or axillary route).
- Any concomitant medications/products/vaccines listed in Section 6.6.2 during the entire study period.
- Any concomitant medication/product/vaccine relevant to a SAE* or administered at any time during the study period for the treatment of a SAE*.

  * SAEs that are required to be reported per protocol.

6.6.2. 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 Section10.4 for study cohorts/ data sets to be analysed.

- Any investigational or non-registered product (drug or vaccine) other than the study vaccines used during the study period.
- Immunosuppressants or other immune-modifying drugs administered chronically (i.e. more than 14 days) during the study period. For corticosteroids, this will mean prednisone ≥ 20 mg/day, or equivalent. 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 from 30 days before the first dose and ending 30 days after the last dose of vaccine with the exception of any influenza vaccine or pneumococcal vaccine (Step 2 only)

which may be administered ≥ 15 days preceding or following any study vaccine dose.*

* In case an emergency mass vaccination for an unforeseen public health threat (e.g. a pandemic) is organised by the public health authorities, outside the routine immunisation programme, 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.

6.7. Intercurrent medical conditions that may lead to elimination of a subject from ATP analyses

At each study visit subsequent to the first vaccination visit, it must be verified if the subject has experienced or is experiencing any intercurrent medical condition(s) requiring medical attention. If it is the case, the condition(s) must be recorded in the eCRF.

Subjects may be eliminated from the ATP cohort for immunogenicity if, during the study, they incur a condition that has the capability of altering their immune response (e.g. confirmed immunosuppressive or immunodeficient condition, including HIV).

7. HEALTH ECONOMICS

Not applicable.

8. 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 will be instructed to contact the investigator immediately should he/she manifest any signs or symptoms they perceive as serious.

8.1. Safety definitions

8.1.1. 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 unfavourable 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:

- Worsening 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 vaccines 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 or a concurrent medication (overdose per se should not be reported as an AE/SAE).
- Signs, symptoms temporally associated with vaccine administration.
- Pre- or post-treatment events that occur as a result of protocol-mandated procedures (i.e. invasive procedures, modification of subject's previous therapeutic regimen).

AEs to be recorded as endpoints (solicited AEs) are described in Section 8.1.3. 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.

8.1.2. 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 hospitalisation or prolongation of existing hospitalisation,

Note: In general, hospitalisation 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 hospitalisation are also considered AEs. If a complication prolongs hospitalisation or fulfils any other serious criteria, the event will also be considered serious. When in doubt as to whether 'hospitalisation' occurred or was necessary, the AE should be considered serious.

Hospitalisation 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, OR

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, diarrhoea, 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.

e. Is a congenital anomaly/birth defect in the offspring of a study subject.

Medical or scientific judgement 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 hospitalisation but may jeopardise 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 hospitalisation.

8.1.3. Solicited adverse events

Solicited local and general AEs occurring during a 7-day follow-up period after each vaccination (i.e. the day of vaccination and the 6 subsequent days), as well as unsolicited AEs occurring during a 30-day follow-up period after each vaccination (i.e. the day of vaccination an the 29 subsequent days), will be reported via paper Diary Cards for vaccine reactogenicity and recorded via the appropriate section of the eCRF.

8.1.3.1. Solicited local (injection-site) adverse events

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

Table 16 Solicited local adverse events

Pain at injection site
Redness at injection site
Swelling at injection site

8.1.3.2. Solicited general adverse events

The following general AEs will be solicited:

Table 17 Solicited general adverse events

Fever
Headache
Fatigue
Myalgia
Gastrointestinal symptoms [a]

[a] Gastrointestinal symptoms include nausea, vomiting, diarrhoea and/or abdominal pain.

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.

8.1.4. 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, haematology, urinalysis) or other abnormal assessments 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 8.1.1 and 8.1.2). 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 judgement in deciding whether an abnormal laboratory finding or other abnormal assessment is clinically significant.

8.1.5. Adverse events of specific interest (potential immune-mediated diseases)

Potential immune-mediated diseases (pIMDs) are a subset of AEs that include autoimmune diseases and other inflammatory and/or neurologic disorders of interest which may or may not have an autoimmune aetiology. AEs that need to be recorded and reported as pIMDs include those listed in Table 18.

However, the investigator will exercise his/her medical and scientific judgement in deciding whether other diseases have an autoimmune origin (i.e. pathophysiology involving systemic or organ-specific pathogenic autoantibodies) and should also be recorded as a pIMD.

Table 18 List of potential immune-mediated diseases

Neuroinflammatory disorders	Musculoskeletal disorders	Skin disorders
• Cranial nerve disorders, including paralyses/paresis (e.g. Bell's palsy) • Optic neuritis • Multiple sclerosis • Transverse myelitis • Guillain-Barré syndrome, including Miller Fisher syndrome and other variants • Acute disseminated encephalomyelitis, including site specific variants: e.g. non-infectious encephalitis, encephalomyelitis, myelitis, myeloradiculoneuritis • Myasthenia gravis, including Lambert-Eaton myasthenic syndrome • Immune-mediated peripheral neuropathies and plexopathies, (including chronic inflammatory demyelinating polyneuropathy, multifocal motor neuropathy and polyneuropathies associated with monoclonal gammopathy). • Narcolepsy	• Systemic lupus erythematosus and associated conditions • Systemic scleroderma (Systemic sclerosis), including diffuse systemic form and CREST syndrome Idiopathic inflammatory myopathies, including dermatomyositis • Polymyositis • Antisynthetase syndrome • Rheumatoid arthritis, and associated conditions including juvenile chronic arthritis and Still's disease • Polymyalgia rheumatica • Spondyloarthritis, including ankylosing spondylitis, reactive arthritis (Reiter's Syndrome) and undifferentiated spondyloarthritis • Psoriatic arthropathy • Relapsing polychondritis • Mixed connective tissue disorder	• Psoriasis • Vitiligo • Erythema nodosum • Autoimmune bullous skin diseases (including pemphigus, pemphigoid and dermatitis herpetiformis) • Alopecia areata • Lichen planus • Sweet's syndrome • Localised Scleroderma (Morphoea)

Vasculitides	Blood disorders	Others
• Large vessels vasculitis including: giant cell arteritis such as Takayasu's arteritis and temporal arteritis. • Medium sized and/or small vessels vasculitis including: polyarteritis nodosa, Kawasaki's disease, microscopic polyangiitis, Wegener's granulomatosis, Churg–Strauss syndrome (allergic granulomatous angiitis), Buerger's disease (thromboangiitis obliterans), necrotizing vasculitis and anti-neutrophil cytoplasmic antibody (ANCA) positive vasculitis (type unspecified), Henoch-Schonlein purpura, Behcet's syndrome, leukocytoclastic vasculitis.	• Autoimmune hemolytic anemia • Autoimmune thrombocytopenia • Antiphospholipid syndrome • Pernicious anemia • Autoimmune aplastic anaemia • Autoimmune neutropenia • Autoimmune pancytopenia	• Autoimmune glomerulonephritis (including IgA nephropathy, glomerulonephritis rapidly progressive, membranous glomerulonephritis, membranoproliferative glomerulonephritis, and mesangioproliferative glomerulonephritis) • Ocular autoimmune diseases (including autoimmune uveitis and autoimmune retinopathy) • Autoimmune myocarditis/cardiomyopathySarcoidosis • Stevens-Johnson syndrome • Sjögren's syndrome • Idiopathic pulmonary fibrosis • Goodpasture syndrome • Raynaud's phenomenon

Liver disorders	Gastrointestinal disorders	Endocrine disorders
• Autoimmune hepatitis • Primary biliary cirrhosis • Primary sclerosing cholangitis • Autoimmune cholangitis	• Inflammatory Bowel disease, including Crohn's disease, ulcerative colitis, microscopic colitis, ulcerative proctitis • Celiac disease • Autoimmune pancreatitis	• Autoimmune thyroiditis (including Hashimoto thyroiditis) • Grave's or Basedow's disease • Diabetes mellitus type I • Addison's disease • Polyglandular autoimmune syndrome • Autoimmune hypophysitis

When there is enough evidence to make any of the above diagnoses, the AE must be reported as a pIMD. Symptoms, signs or conditions which might (or might not) represent the above diagnoses, should be recorded and reported as AEs but not as pIMDs until the final or definitive diagnosis has been determined, and alternative diagnoses have been eliminated or shown to be less likely.

In order to facilitate the documentation of pIMDs in the eCRF, a pIMD standard questionnaire and a list of preferred terms (PTs) and PT codes corresponding to the above diagnoses will be available to investigators at study start.

8.2. Events or outcomes not qualifying as adverse events or serious adverse events

Pregnancy

Female subjects who are pregnant or lactating at the time of vaccination must not receive additional doses of study vaccines but may continue other study procedures at the discretion of the investigator.

While pregnancy itself is not considered an AE or SAE, any adverse pregnancy outcome or complication or elective termination of a pregnancy for medical reasons will be recorded and reported as an AE or a SAE.

Note: The pregnancy itself should always be recorded on an electronic pregnancy report.

The following should always be considered as SAE and will be reported as described in Sections 8.4.1 and 8.4.3:

- Spontaneous pregnancy loss, including:
  - spontaneous abortion, (spontaneous pregnancy loss before/at 22 weeks of gestation)
  - ectopic and molar pregnancy
  - stillbirth (intrauterine death of foetus after 22 weeks of gestation).

  Note: the 22 weeks cut-off in gestational age is based on WHO-ICD 10 noted in the EMA Guideline on pregnancy exposure [EMA, 2006]. It is recognized that national regulations might be different.
- Any early neonatal death (i.e. death of a live born infant occurring within the first 7 days of life).

- Any congenital anomaly or birth defect (as per [CDC MACDP] guidelines) identified in the offspring of a study subject (either during pregnancy, at birth or later) regardless of whether the foetus is delivered dead or alive. This includes anomalies identified by prenatal ultrasound, amniocentesis or examination of the products of conception after elective or spontaneous abortion.

Furthermore, any SAE occurring as a result of a post-study pregnancy AND considered by the investigator to be reasonably related to the investigational vaccines will be reported to GSK Biologicals as described in Section 8.4.3. While the investigator is not obligated to actively seek this information from former study participants, he/she may learn of a pregnancy through spontaneous reporting.

8.3. Detecting and recording adverse events, serious adverse events and pregnancies

8.3.1. Time period for detecting and recording adverse events, serious adverse events and pregnancies

All AEs starting within 30 days following administration of each dose/ of study vaccines (Day 0 to Day 30) 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 vaccines and will end at study conclusion. See Section 8.4 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 until study conclusion.

In addition to the above-mentioned reporting requirements and in order to fulfil 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 and recording of pIMDs will begin at the first receipt of study vaccine and will end at study conclusion. See Section 8.4 for instructions on reporting of pIMDs.

The time period for collecting and recording pregnancies will begin at the first receipt of study vaccine and will end at study conclusion. See Section 8.4 for instructions on reporting of pregnancies.

The time period for collecting and recording of intercurrent medical conditions requiring medical attention will begin at the first receipt of study vaccine and will end at study conclusion.

Table 19 Reporting periods for collecting safety information

Event	Screening Visit*	Visit 1 Dose 1 Day 0	7 d post Dose 1 Day 6	30 d post Dose 1 Day 29	Visit 4 Dose 2 Day 60	7 d post Dose 2 Day 66	30 d post Dose 2 Day 89	Day 210	Study Conclusion Day 420
Solicited local and general AEs									
Unsolicited AEs									
AEs/SAEs leading to withdrawal from the study									
SAEs									
SAEs related to study participation or concurrent GSK medication/vaccine									
Pregnancies									
pIMDs									
Intercurrent medical condictions requiring medical attention									

d = day follow-up period.
* i.e. consent obtained.
The double-bordered lines indicate timings of vaccination.

8.3.2. 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 19. 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.

8.3.3. Evaluation of adverse events and serious adverse events

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

As a consistent method of collecting AEs, the subject should be asked a non-leading question such as:

'*Have you felt different in any way since receiving the vaccine or since the previous 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.

8.3.3.2. Assessment of adverse events

8.3.3.2.1. Assessment of intensity

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

Table 20 Intensity scales for solicited symptoms

Adverse Event	Intensity grade	Parameter
Pain at injection site	0	None
	1	Mild: Any pain neither interfering with nor preventing normal every day activities.
	2	Moderate: Painful when limb is moved and interferes with every day activities.
	3	Severe: Significant pain at rest. Prevents normal every day activities.
Redness at injection site		Record greatest surface diameter in mm
Swelling at injection site		Record greatest surface diameter in mm
Fever[a]		Record temperature in °C
Headache	0	Normal
	1	Mild: Headache that is easily tolerated
	2	Moderate: Headache that interferes with normal activity
	3	Severe: Headache that prevents normal activity
Fatigue	0	Normal
	1	Mild: Fatigue that is easily tolerated
	2	Moderate: Fatigue that interferes with normal activity
	3	Severe: Fatigue that prevents normal activity
Myalgia[b]	0	None
	1	Mild: Myalgia that is easily tolerated
	2	Moderate: Myalgia that interferes with normal activity
	3	Severe: Myalgia that prevents normal activity
Gastrointestinal symptoms (nausea, vomiting, diarrhoea and/or abdominal pain)	0	Gastrointestinal symptoms normal
	1	Mild: Gastrointestinal symptoms that are easily tolerated
	2	Moderate: Gastrointestinal symptoms that interfere with normal activity
	3	Severe: Gastrointestinal symptoms that prevent normal activity

[a] Fever is defined as a temperature ≥ 37.5°C for oral or axillary route. The preferred route for recording temperature in this study will be oral.

[b] Myalgia is defined as generalised muscle pain with an intensity judged by the subject and confirmed by the investigator as more severe than expected for the subject's condition or higher than pre-vaccination.

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

0 : ≤ 20 mm
1 : > 20 mm to ≤ 50 mm
2 : > 50 mm to ≤ 100 mm
3 : > 100 mm

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

0	:	< 37.5 °C
1	:	≥ 37.5 °C to ≤ 38.5 °C
2	:	> 38.5 °C to ≤ 39.5°C
3	:	> 39.5°C

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 judgement.

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 work/school and would necessitate the administration of corrective therapy.

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 8.1.2.

8.3.3.2.2. Assessment of causality

The investigator is obligated to assess the relationship between investigational vaccines and the occurrence of each AE/SAE. The investigator will use clinical judgement 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.

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

If an event meets the criteria to be determined as 'serious' (see Section 8.1.2), 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).

8.3.3.3. 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).

8.3.3.4. Medically attended visits

For each solicited and unsolicited symptom the subject experiences, the subject will be asked if he/she received medical attention defined as hospitalisation, 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.

8.4. Reporting of serious adverse events, pregnancies, and other events

8.4.1. Prompt reporting of serious adverse events, pregnancies, and other events to GSK Biologicals

SAEs that occur in the time period defined in Section 8.3 will be reported promptly to GSK within the timeframes described in Table 21, once the investigator determines that the event meets the protocol definition of a SAE.

pIMDs that occur in the time period defined in Section 8.3 will be reported promptly to GSK within the timeframes described in Table 21, once the investigator determines that the event meets the protocol definition of a pIMD.

Pregnancies that occur in the time period defined in Section 8.3 will be reported promptly to GSK within the timeframes described in Table 21, once the investigator becomes aware of the pregnancy.

Table 21 Timeframes for submitting serious adverse event, pregnancy and other events 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
pIMDs	24 hours**‡	electronic Expedited Adverse Events Report	24 hours*	electronic Expedited Adverse Events Report
Pregnancies	2 weeks*	electronic pregnancy report	2 weeks*	electronic pregnancy report

* Timeframe allowed after receipt or awareness of the information.
**Timeframe allowed once the investigator determines that the event meets the protocol definition of a pIMD.
‡ The investigator will be required to confirm review of the SAE/pIMD causality by ticking the 'reviewed' box in the electronic Expedited Adverse Events Report within 72 hours of submission of the SAE/pIMD.

8.4.2. Contact information for reporting serious adverse events, pregnancies and pIMDs

Study Contact for Reporting SAEs, pIMDs and pregnancies
Refer to the local study contact information document.
Back-up Study Contact for Reporting SAEs, pIMDs and pregnancies
24/24 hour and 7/7 day availability: **GSK Biologicals Clinical Safety & Pharmacovigilance** Fax: PPD or PPD Email address: Rix.CT-safety-vac@gsk.com

8.4.3. 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.

8.4.3.1. 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.

8.4.4. Reporting of pIMDs to GSK Biologicals

Once a pIMD is diagnosed (serious or non-serious) in a study subject, the investigator (or designate) must complete the information in the electronic Expedited Adverse Events Report WITHIN 24 HOURS after he/she becomes aware of the diagnosis. The report allows to specify that the event is a pIMD and whether it is serious or non-serious. The report will always be completed as thoroughly as possible with all available details of the

event, in accordance with the pIMD standard questionnaire provided. Even if the investigator does not have all information regarding a pIMD, 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 pIMD causality by ticking the 'reviewed' box in the electronic Expedited Adverse Events Report within 72 hours of submission of the pIMD.

Refer to Section 8.4.3.1 for back-up system in case the electronic reporting system does not work.

8.4.5. Completion and transmission of pregnancy reports to GSK Biologicals

Once the investigator becomes aware that a subject is pregnant, the investigator (or designate) must complete the required information onto the electronic pregnancy report WITHIN 2 WEEKS.

Note: Conventionally, the estimated gestational age (EGA) of a pregnancy is dated from the first day of the last menstrual period (LMP) of the cycle in which a woman conceives. If the LMP is uncertain or unknown, dating of EGA and the estimated date of delivery (EDD) should be estimated by ultrasound examination and recorded in the pregnancy report.

8.4.6. Updating of SAE, pregnancy, and pIMD information after removal of write access to the subject's eCRF

When additional SAE, pIMD, or pregnancyinformation 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 21.

8.4.7. Regulatory reporting requirements for serious adverse events

The investigator will promptly report all SAEs to GSK in accordance with the procedures detailed in Section 8.4.1. 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 fulfil specific regulatory and GCP requirements, regarding the product under investigation.

8.5. Follow-up of adverse events, serious adverse events, and pregnancies

8.5.1. Follow-up of adverse events and serious adverse events

8.5.1.1. Follow-up during the study

After the initial AE/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 SAEs; refer to Table 21).

All SAEs and pIMDs (serious or non-serious) 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 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.

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

The investigator will follow subjects with SAEs, pIMDs (serious or non-serious), or subjects withdrawn from the study as a result of an AE, until the event has resolved, subsided, stabilised, 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 a paper/ electronic Expedited Adverse Events Report and/or pregnancy report as applicable.

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 recognised follow-up period, GSK Biologicals will be provided with any available post-mortem findings, including histopathology.

8.5.2. Follow-up of pregnancies

Pregnant subjects will be followed to determine the outcome of the pregnancy. At the end of the pregnancy, whether full-term or premature, information on the status of the mother and child will be forwarded to GSK Biologicals using the electronic pregnancy report and the Expedited Adverse Events Report if applicable. Generally, the follow-up period doesn't need to be longer than six to eight weeks after the estimated date of delivery.

Regardless of the reporting period for SAEs for this study, if the pregnancy outcome is a SAE, it should always be reported as SAE.

8.6. 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 6.6).

8.7. 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 8.4.1).

8.8. 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 study 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 the automated Internet-based system).

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: PPD (GSK Biologicals Central Safety Physician on-call) **GSK Biologicals' Central Safety Physician Back-up:** PPD **Emergency Unblinding Documentation Form transmission:** Fax: PPD or PPD

8.9. Subject card

Study subjects 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. 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 must be instructed to keep subject cards in their possession at all times.

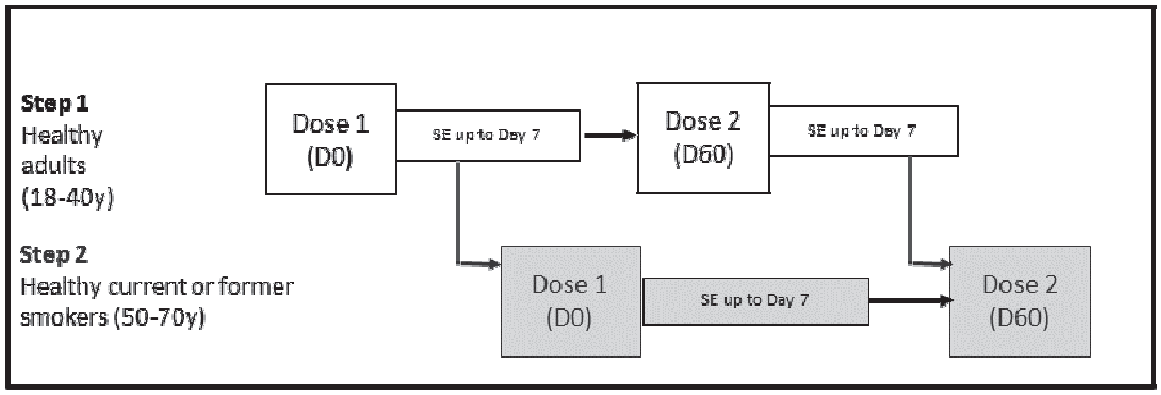
8.10. Holding rules and safety monitoring

The investigator is not permitted to start the administration of the next dose (Dose2) or next step (Step 2) until receipt of the favourable outcome of the safety evaluation, based on the safety data collected up to seven days after administration of the previous dose(s) (Figure 2).

In order to move to the next step, safety data will be needed from all subjects. In order to move from Dose 1 to Dose 2 within a step, safety data from about 50% of the vaccinated subjects will be needed (Figure 2).

Moreover, if the investigator becomes aware of a holding rule being met, he/she will suspend vaccination and will inform GSK Biologicals Central Safety immediately (within 24 hours) (e.g. in case of SAEs).

Figure 2 Overview of iSRC evaluations



D = day
SE up to Day 7 = safety evaluation based on data up to the Day 7 post-vaccination visit (*i.e.* Visit 2 at Day 7 and Visit 5 at Day 67), including Day 7 post-vaccination laboratory assessments.

8.10.1. Safety Review Team

The project's SRT includes as core members the GSK Biologicals' Central Safety Leader, the Clinical Research & Development Lead (CRDL), Epidemiologist, Global Regulatory Lead and a Biostatistician of the project. The SRT is responsible for on-going safety monitoring of the entire project and meets on a regular basis. The SRT will inform the iSRC about any potential safety concern relevant to the study.

Before each iSRC safety evaluation in this study (see below), the SRT will review the same safety data, but in a *blinded* manner in order to keep all people involved in the conduct, cleaning and final analysis of the study blinded.

8.10.2. Internal Safety Review Committee

As the investigational vaccine formulation will be administered for first time to human, an iSRC will be appointed in addition to the existing project's SRT and safety holding rules have been defined.

The iSRC, authorised by the GSK Biologicals' Vaccine Safety Monitoring Board (VSMB) will include as core members a GSK Biologicals' Safety Leader, a CRDL and a Biostatistician, who are not otherwise involved in the conduct of the project, in order to maintain the observer-blind to the treatment codes amongst project-related personnel until the final analysis of data.

The iSRC will conduct *unblinded* reviews of all available safety data from the present study, while taking into account any other findings that could have an impact on the safety of the subjects, and will determine whether there is a safety signal that needs to be escalated to GSK Biologicals' VSMB. In the event that a safety signal is observed, GSK Biologicals' VSMB might decide to suspend, modify or continue the conduct of the study in all groups or in selected groups.

To facilitate the safety evaluation, the investigator or his/her designee will need to transcribe the information of the Diary Cards returned at the 7 day post-vaccination visits into the eCRF promptly (preferably within two working days of data availability).

8.10.2.1. Outcome of safety evaluation

If **no safety signal** is observed, the favourable outcome of the safety evaluation will be documented and provided in writing (scanned and emailed) authorising the investigator to proceed to the next dose and/or step of the trial.

If a **safety signal** is observed during the safety evaluation or if any of the holding rules 1a, 1b, 1c is reported by the investigator at any time, the SRT and iSRC leader is responsible for the urgent communication and escalation of the concern to the GSK Biologicals VSMB who will decide during an *ad hoc* meeting whether to suspend, modify or continue the conduct of the study on all groups or on selected groups. The decision of the GSK Biologicals VSMB will be documented and provided in writing (scanned and emailed) to the investigator regarding the further conduct of the study.

In the event that a safety signal has prompted a GSK Biologicals' VSMB review, further vaccination may resume following, and dependent upon, a decision provided in writing to the investigator (scanned and emailed) authorising the resumption of vaccinations.

8.10.2.2. Process for the suspension of vaccination and/or study modification

In the event that a safety signal is observed, GSK Biologicals' VSMB might decide to suspend further vaccinations in all groups or in selected groups.

In case the vaccination is suspended:

- Subjects who signed an informed consent but have not received any study vaccine will be informed that their study participation will be stopped.
- For all other subjects, all visits will continue as planned for evaluation of the safety and immunogenicity of the vaccine except that Visit 5 on Day 67 will be cancelled if no vaccine dose is administered at Visit 4 on Day 60.

8.10.3. Study holding rules

The safety holding rules which will be checked by the iSRC during each safety evaluation are defined below (Table 22). Please refer to APPENDIX C for the definition of the Food and Drug Administration (FDA) Toxicology Grading Scale for laboratory abnormalities in healthy subjects.

These holding rules have been written under the assumption that the safety data will be available from all subjects. If the data from all subjects are not available (i.e. in case a subject is lost to follow-up), then the holding rules will be assessed on a pro-rata basis.

Table 22 List of holding rules

Holding rule N°	Event	Number of subjects	
		Step 1	Step 2
1a	Death or any life-threatening SAE	≥ 1	≥ 1
1b	Any **withdrawal** from the study (by investigator or subject request) following a Grade 3 AE that cannot reasonably be attributed to a cause other than vaccination	≥ 1	≥ 1
1c	Any local or general solicited AE leading to **hospitalisation**, or **fever >40°C** (104°F) that cannot reasonably be attributed to a cause other than vaccination, or **necrosis** at the injection site, within the 7-day (Day 0-Day 6) post-vaccination period	≥ 1	≥ 1
2a	Any **Grade 3 solicited local** AE lasting 48h or more in an investigational group, within the 7-day (Day 0-Day 6) post-vaccination period	≥ 5/15 and ≥ 2 subjects/group	≥ 8/30 and ≥ 2 subjects/group
2b	Any **Grade 3 solicited general** AE lasting 48h or more in an investigational group, that cannot reasonably be attributed to a cause other than vaccination, within the 7-day (Day 0-Day 6) post-vaccination period	≥ 5/15 and ≥ 2 subjects/group	≥ 8/30 and ≥ 2 subjects/group
2c	Any **Grade 3 unsolicited** AE in an investigational group, that cannot reasonably be attributed to a cause other than vaccination, within the 7-day (Day 0-Day 6) post-vaccination period or Any Grade 3 abnormality in pre-specified haematological or biochemical **laboratory parameters*** in an investigational group within the 7-day (Day 0-Day 6) post-vaccination period	≥ 5/15 and ≥ 2 subjects/group	≥ 8/30 and ≥ 2 subjects/group

* i.e. haemoglobin, WBC, platelets, creatinine, ALT and AST

8.10.4. Risk assessments

Figure 3 gives the probability of not meeting each holding rule 1 and 2 for the 15 subjects in a candidate vaccine group (as in Step 1 of this study). The figure illustrates that holding rule 1 has more than 86% chance of not being met for an event with a true incidence rate below 1.0% and has less than 3.5% of chance of not being met for an event with a true incidence rate above 20%.

Figure 3 **Risk assessment curve for safety holding rules 1 and 2 for treatment arms of 15 subjects (no adjustment for multiplicity)**

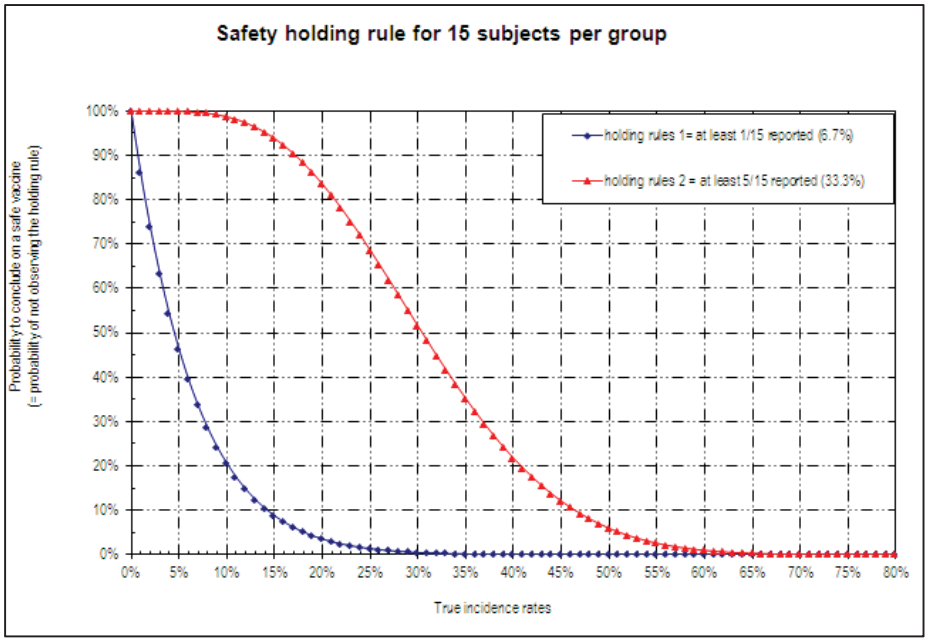
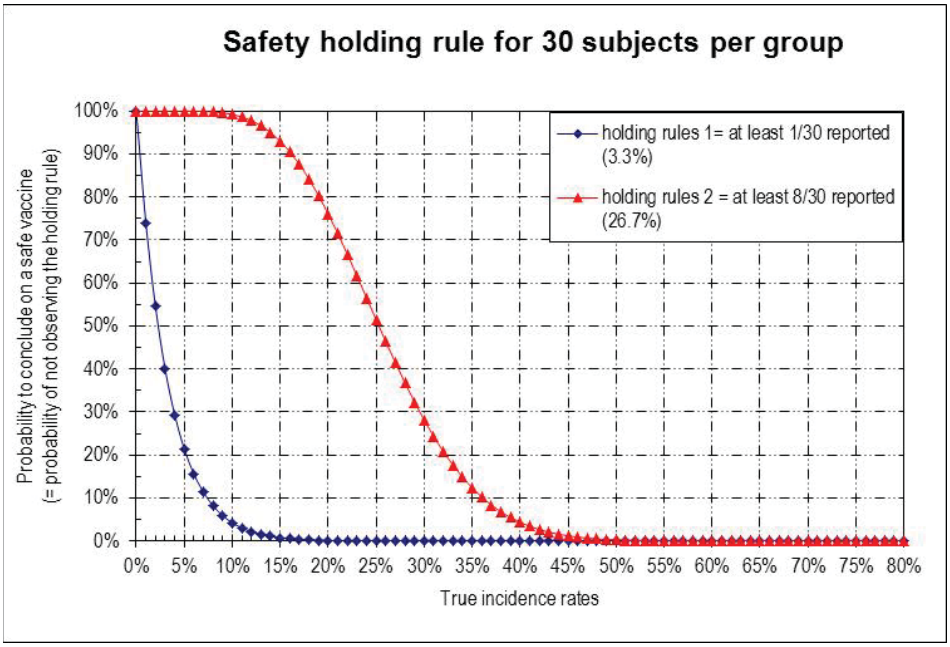


Figure 4 gives the probability of not meeting holding rules 1 and 2 for the 30 subjects in a candidate vaccine group (as in Step 2 of this study). The figure illustrates that holding rule 1 has more than 74% chance of not being met for an event with a true incidence rate below 1% and has less than 4.2% of not being met for an event with a true incidence rate above 10%.

Figure 4 **Risk assessment curve for safety holding rules 1 and 2 for treatment arms of 30 subjects (no adjustment for multiplicity)**



9. SUBJECT COMPLETION AND WITHDRAWAL

9.1. Subject completion

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

9.2. Subject withdrawal

Withdrawals will not be replaced.

9.2.1. 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/was not available for the concluding contact 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 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 himself/herself, or by the investigator, as well as which of the following possible reasons was responsible for withdrawal:

- SAE.
- Non-serious AE.
- 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 he/she has withdrawn consent, the investigator will document the reason for withdrawal of consent, if specified by the subject, 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 8.5.1.2).

9.2.2. 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.

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

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

9.3. Screening 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.
- SAEs related to study participation, to concomitant use of GSK products or any fatal SAEs.
- Screening conclusion.

10. STATISTICAL METHODS

10.1. Primary endpoints

- Occurrence of each solicited local and general AE, during a 7-day follow-up period (*i.e.* day of vaccination and 6 subsequent days) post-Dose 1 and post-Dose 2, in all subjects, in all vaccine groups.
- Occurrence of any unsolicited AEs, during a 30-day follow-up period (i.e. day of vaccination and 29 subsequent days) post-Dose 1 and post-Dose 2, in all subjects, in all vaccine groups.

- Occurrence of haematological and biochemical laboratory abnormalities, after vaccination, in all subjects, in all vaccine groups:
  - Any haematological (RBC, WBC and differential count, platelets count and haemoglobin level) or biochemical (ALT, AST and creatinine) laboratory abnormality on Day 7, Day 60, Day 67, Day 210 and Day 420.
- Occurrence of any SAE, occurring from first vaccination (Day 0) to study conclusion (Day 420) in all subjects, in all vaccine groups.
- Occurrence of any pIMD occurring from first vaccination (Day 0) to study conclusion (Day 420) in all subjects, in all vaccine groups.

10.2. Secondary endpoints

- Humoral immune response to the components of the NTHi-Mcat vaccine formulations, on Day 0, Day 30, Day 60, Day 90, Day 210 and Day 420, in all subjects, in all vaccine groups:
  - Anti-PD, anti-PE, anti-PilA and anti-UspA2 antibody concentrations.
- Cell-mediated immune response to components of the NTHi-Mcat vaccine formulations, on Day 0, Day 60, Day 90, Day 210 and Day 420, in a sub-cohort of subjects, in all vaccine groups:
  - Frequency of specific $CD4^+/CD8^+$ T-cells measured on cryopreserved peripheral blood mononuclear cells (PBMCs) and identified by flow cytometry intracellular cytokine staining (ICS) expressing two or more markers (such as IL-2, IL-13, IL-17, IFN-γ, TNF-α and CD40L).

10.3. Determination of sample size

This study is a Phase I safety study to get a preliminary safety assessment of the vaccine formulations in healthy adults.

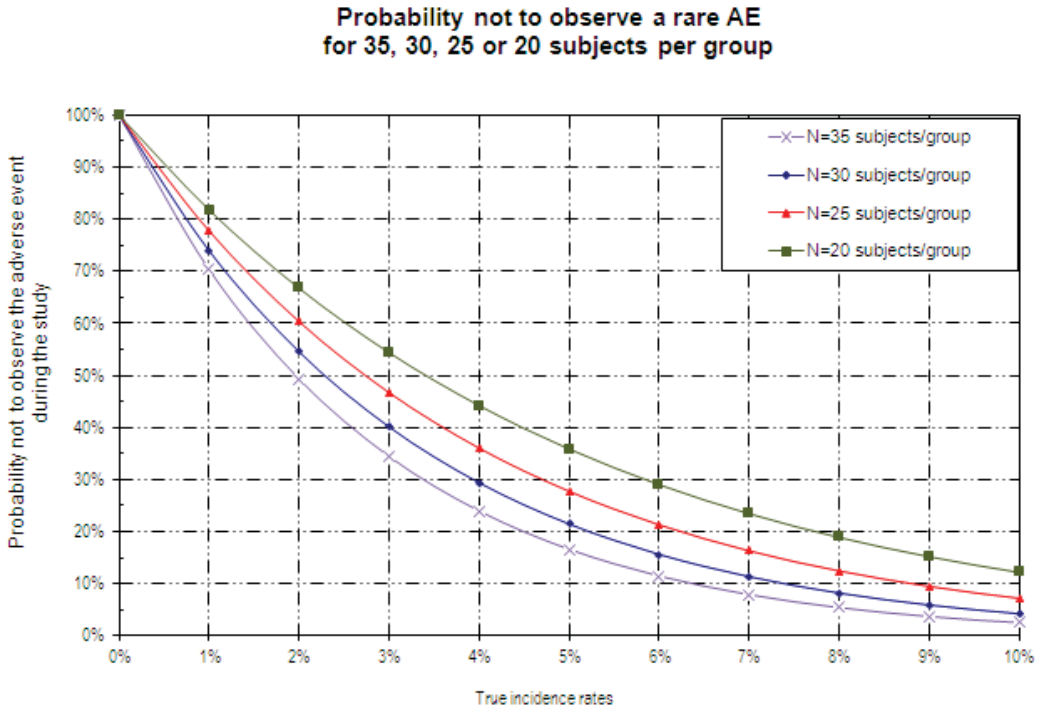
Considering the sample size of 15 in each study group in Step 1, Table 23 illustrates the precision one can get on the percentage of subjects with symptoms following vaccination.

Table 23 Exact confidence interval for one proportion with 15 subjects

Number(%) of subjects with a symptom	Exact 95% Confidence Interval (CI)	
	Lower Limit	Upper limit
0 (0.0)	0.0	21.8
1 (6.7)	0.2	31.9
2 (13.3)	1.7	40.5
3 (20.0)	4.3	48.1
4 (26.7)	7.8	55.1
5 (33.3)	11.8	61.6
6 (40.0)	16.3	67.7
7 (46.7)	21.3	73.4
8 (53.3)	26.6	78.7
9 (60.0)	32.3	83.7
10 (66.7)	38.4	88.2
11 (73.3)	44.9	92.2
12 (80.0)	51.9	95.7
13 (86.7)	59.5	98.3
14 (93.3)	68.1	99.8
15 (100.0)	78.2	100

Considering the sample size of 30 or less in each study group in Step 2, Figure 5 illustrates the probability of detecting an AE with a true occurrence rate of 0 - 10%.

Figure 5 Probability of not detecting an AE with a true occurrence rate of 0-10%



The probability of detecting an AE with a true incidence of 5% is around 78.5% with 30 subjects per group.

10.4. Cohorts for analyses

The following subject cohorts will be evaluated.

10.4.1. Total vaccinated cohort

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

10.4.2. According-to-protocol cohort

The ATP cohorts will be defined by epoch and will consist of all subjects from the TVC who will comply with eligibility criteria, study procedures up to the end of the epoch and had immunogenicity results in the epoch.

10.4.2.1. According-to-protocol cohort for analysis of immunogenicity (Epoch 001)

The ATP cohort for immunogenicity will include all subjects in the TVC:

- Who met all eligibility criteria.
- For whom the administration route and site of the vaccines was according to protocol.
- Who complied with the vaccination schedule, as specified in Table 7.
- Who did not receive a concomitant medication/ product/vaccine leading to the elimination from the ATP analysis (see Section 6.6.2) up the one month post-Dose 2 visit (Visit 6, Day 90).
- Who did not present with an intercurrent medical condition leading to elimination from the ATP analysis (see Section 6.7) up to the one month post-Dose 2 visit (Visit 6, Day 90).
- Who complied with the immunogenicity blood sample timings as specified in Table 7 at 1 month post-Dose 2 visit (Visit 6, Day 90).
- For whom post-Dose 2 immunogenicity results are available for at least one assay.

10.4.2.2. According-to-protocol cohort for analysis of persistence of immunogenicity (Epoch 002)

The ATP cohort for persistence of immunogenicity will include all evaluable subjects, i.e., those who were included in the ATP cohort for immunogenicity, or were excluded from this cohort solely because they had no blood samples taken or because of non-compliance with blood sample timings up to the one month post-Dose 2 visit (Visit 6, Day 90), and:

- Who did not receive a concomitant medication/ product/vaccine leading to elimination from the ATP analysis for immunogenicity (see Section 6.6.2).
- Who did not present with an intercurrent medical condition leading to elimination from the ATP analysis for immunogenicity (see Section 6.7).
- Who complied with at least one of the blood sample timings after the one month post-last vaccination visit (Visit 6, Day 90) as specified in Table 7.
- For whom persistence immunogenicity results are available for at least one assay in at least one of the two persistence time points (Visit 7, Day 210 and Visit 8, Day 420).

10.5. Derived and transformed data

The study groups will be defined by treatment actually administered at Dose 1.

Demography

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

Immunogenicity

- For a given subject and the analysis of a given immunogenicity measurement, missing or un-evaluable measurements will not be replaced.
- A seronegative subject is defined as a subject whose antibody concentration is below the assay cut-off value.
- A seropositive subject is defined as a subject whose antibody concentration is greater than or equal to the assay cut-off value.
- Antibody concentrations below the assay cut-off will be given an arbitrary value of half the assay cut-off for the purpose of geometric mean concentration (GMC) calculation.
- Calculation of the GMCs will be performed by taking the anti-logarithm in base 10 (anti-log10) of the mean of the log10 concentration/titre transformations.
- All confidence intervals (CIs) computed will be two-sided 95% CIs.

Safety/reactogenicity

- For **solicited symptoms**, the analysis will exclude subjects with missing or un-evaluable measurements (e.g. total analysis of solicited symptoms will include all vaccinated subjects with documented solicited symptom sheets).
- For the **unsolicited symptoms** and **concomitant medications/ products/ vaccinations**, subjects who did not report unsolicited symptoms/concomitant medications/ products/ vaccinations will be treated as subjects without unsolicited symptoms or concomitant medications/ products/ vaccinations, respectively.

10.6. Statistical analysis

The final analyses will be descriptive and will be presented by study group.

10.6.1. Analysis of demographics

- Demographic characteristics (age at the first dose in years, gender and race), cohort description, withdrawal status will be summarised by group using descriptive statistics:
  - Frequency tables will be generated for categorical variables such as race;
  - Mean, median and standard error will be provided for continuous data such as age.
- The distribution of subjects enrolled among the study sites will be tabulated as a whole and per group.
- Withdrawal status will be summarised by group using descriptive statistics:
  - The numbers of withdrawn subjects will be tabulated according to the reason for withdrawal.
- The number of subjects enrolled into the study as well as the number of subjects excluded from ATP analyses will be tabulated.

10.6.2. Analysis of safety

The primary analysis will be performed on the TVC and, if in any vaccine group and at any time point the percentage of vaccinated subjects excluded from the ATP cohort is at least 10%, a second analysis will be performed on the ATP cohort to complement 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 after Dose 1 and Dose 2 and overall with exact 95% CI. The same computations will be done for Grade 3 AEs, any AEs causally related to vaccination and any Grade 3 AEs causally related to vaccination.

The percentage of subjects/doses reporting each individual solicited local (any grade, Grade 3) and general (any grade, Grade 3, any causally related to vaccination and any

Grade 3 causally related to vaccination) AE during the 7-days (Day 0 to Day 6) follow-up period will be tabulated for each group as follows:

- Overall, the percentage of subjects with the symptom and its exact 95% CI.
- Overall, the percentage of doses with the symptom and its exact 95% CI.
- At each study dose (visit), the percentage of subjects with the symptom and its exact 95% CI.

The exact 95% CIs will be calculated assuming independence between doses. For fever, additional analyses will be performed by 0.5°C increments.

The verbatim reports of **unsolicited symptoms** will be reviewed by a physician and the signs and symptoms will be coded according to the MedDRA Dictionary for Adverse Reaction Terminology. Every verbatim term will be matched with the appropriate preferred term (PT). The percentage of subjects with unsolicited symptoms within 30 days after any doses with its exact 95% CI will be tabulated by group and by MedDRA PT. Similar tabulation will be done for Grade 3 unsolicited symptoms, for unsolicited symptoms that resulted in a medically attended visit, for unsolicited symptoms causally related to vaccination and for Grade 3 symptoms causally related to the vaccination.

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

- The percentage of subjects having haematology and biochemistry results below or above the normal laboratory ranges will be tabulated by time point.
- The maximum grading from Screening up to Visit 8 (Day 420) will be tabulated (grades will be based on local laboratory normal ranges and derived from FDA Guidance to Industry "Toxicity Grading Scale for Healthy Adult and Adolescent Volunteers Enrolled in Preventive Vaccine Clinical Trials", see APPENDIX C. Those laboratory parameters not included in the FDA grading scale will not be graded).

The number of subjects who experienced at least one **SAE** or any **pIMDs** during the entire study period will be reported.

The percentage of subjects/dose using **concomitant medication/ product** (any medication/ product, any antipyretic and any antipyretic taken prophylactically, respectively) during the 30-day follow-up period (Day 0 – Day 29) will be summarised per group for each dose and overall per dose.

The number of subjects who experienced any **AE leading to study withdrawal**, from first vaccination up to study conclusion, or any **SAE related to study participation of concurrent GSK medication/ vaccination,** during the entire study period, will be reported.

iSRC safety evaluations

At each iSRC time point, a blinded review will be done by the GSK SRT followed by an unblinded review of safety data by the iSRC (refer to Section 8.10 for more information). No individual clinical study report (CSR) will be written as a result of these safety evaluations.

10.6.3. Analysis of immunogenicity

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

10.6.3.1. Humoral immune response

Within group evaluation

For each group, at each time point during which blood samples are collected for humoral immune response and for each assay:

- Seropositivity rate and their exact 95% CI will be tabulated.
- GMCs and their 95% CI will be calculated.
- Antibody concentrations distribution will be investigated using Reverse Cumulative Curves.

These analyses will also be performed for each level of the following minimisation factors: age (50-59 years vs. 60-70 years), smoking status (current vs. former smoker), FEV_1/FCV ratio (≥0.7 vs. <0.7).

Between groups evaluation (only for Step 2)

Comparative analyses will be exploratory with the aim to characterise the difference between the 10-10-10-AS and 10-10-3-AS groups in humoral immune response.

The difference in terms of GMCs will be evaluated, one month post-Dose 2, by computing the 95% CIs of the GMC ratio between groups for Step 2 by using a one-way ANCOVA model on the logarithm10 transformation of the concentrations/ titres. The ANCOVA model will include the group category, the age category, the smoking status and FEV_1/FVC (≥ 0.7 or < 0.7) ***and the pre-Dose 1 concentration (as covariate)*** as fixed effects. The groups will be considered significantly different if the 95% CI for the GMC ratio between the two groups does not contain the value 1. **(Amended 16 March 2016)**

However, these differences should be interpreted with caution considering that there will be no adjustment for multiplicity of endpoints.

10.6.3.2. CMI response

The frequency of specific $CD4^+$/$CD8^+$ T-cells will be summarised (descriptive statistics) by group in Step 2 at each time point during which blood samples are collected for CMI.

10.6.3.3. Summary score to support dose selection

For each group in Step 2, a summary score at one month post-Dose 2 (Day 90) will summarise how well the group is performing in terms of immunogenicity and CMI results. The calculation of a summary score for both active groups (i.e. 10-10-10-AS and 10-10-3-AS) will be proposed in order to help the formulation selection as a higher summary score indicates a higher global immunogenicity and CMI response.

The summary scores will be calculated based on the subjects from the ATP cohort.

The calculation of these summary scores from immunogenicity data for the interim, and from immunogenicity data and CMI data for the final analyses, and the strategy for their use in the dose selection will be presented more in detail in the Statistical Analysis Plan.

10.7. Interpretation of analyses

Comparative analyses with the aim to characterise the difference in reactogenicity/ immunogenicity between groups will be descriptive. These descriptive analyses should be interpreted with caution as neither the power or type I error are controlled for.

10.8. 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.

10.8.1. Sequence of analyses

- The analysis of data up to Visit 6 (Day 90, end of Epoch 001) will be performed in a first step. This analysis will include:
  - The final analysis of all immunogenicity results (including CMI), solicited AEs post-Dose 1 and post-Dose 2,
  - The assessment of unsolicited AEs up to 30 days post-Dose 1 and post-Dose 2, and of SAEs and pIMDs up to 30 days post-Dose 2 on as cleaned as possible data. No individual listings will be written at this stage.

  This analysis will be documented in a statistical report. At this point, the GSK statistician will be unblinded (i.e. will have access to the individual subject treatment assignments).

- The analysis of Epoch 002 (from Visit 7 [Day 210] up to Visit 8 [Day 420]) will be performed in a second step, once those data will be available and cleaned. This analysis will include:
  - The final analysis of all immunogenicity results (including CMI) up to Visit 8 (Day 420).
  - SAEs and pIMDs up to Visit 8 (Day 420) on cleaned data.

  In addition, all previous analyses will be re-produced based on cleaned data at this point.

  Individual listings will only be provided at this stage.

All study results will be presented in an integrated study report at the end of the study. **(Amended 16 March 2016)**

10.8.2. Statistical considerations for interim analyses

No adjustment for multiplicity will be performed for the comparative analyses since they are all descriptive and will all be performed on final data.

11. ADMINISTRATIVE MATTERS

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

11.1. 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.

11.2. 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 (SDV). By SDV 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.

11.3. 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.

11.4. 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.

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

Study information from this protocol will be posted on publicly available clinical trial registers before enrolment of subjects begins.

Summaries of the results of GSK interventional studies (phase I-IV) are posted on publicly available results registers within 12 months of the primary completion date for studies of authorised vaccines and 18 months for studies of non-authorised vaccines.

GSK also aims to publish the results of these studies in the searchable, peer reviewed scientific literature. Manuscripts are submitted for publication within 24 months of the last subject's last visit. At the time of publication, this protocol will be fully disclosed.

11.6. 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.

12. COUNTRY SPECIFIC REQUIREMENTS

Not applicable.

13. REFERENCES

Alamoudi OS. Bacterial infection and risk factors in outpatients with acute exacerbation of chronic obstructive pulmonary disease: a 2-year prospective study. *Respirology* 2007; 12:283-7.

American Thoracic Society and European Respiratory Society. Standards for the diagnosis and management of patients with COPD. 2004. New York, USA; Lausanne, Switzerland. Ref Type: Conference.

Attia AS, Lafontaine ER, Latimer JL, *et al.* The UspA2 protein of *Moraxella catarrhalis* is directly involved in the expression of serum resistance. *Infect Immun* 2005;73(4):2400-2410.

Bandi V, Jakubowycz M, Kinyon C, *et al*. Infectious exacerbations of chronic obstructive pulmonary disease associated with respiratory viruses and non-typeable *Haemophilus influenzae*. *FEMS Immunol Med Microbiol*. 2003;37:69-75.

Barcelo B, Pons J, Ferrer JM, *et al*. Phenotypic characterisation of T-lymphocytes in COPD: abnormal CD4+CD25+ regulatory T-lymphocyte response to tobacco smoking. *Eur Respir J* 2008;31:555-562.

Buist S, McBurnie MA, Vollmer WM, *et al*. International variation in the prevalence of COPD (The BOLD Study): a population-based prevalence study. *Lancet* 2007;370:741-50.

Cazzola M, MacNee W, Martinez FJ, *et al*. Outcomes for COPD pharmacological trials: from lung function to biomarkers. *Eur Respir J* 2008;31:416-69.

Centers for Disease Control and Prevention Metropolitan Atlanta Congenital Defects Program (CDC MACDP) guidelines. Birth defects and genetic diseases branch 6-digit code for reportable congenital anomalies; *http://www.cdc.gov/ncbddd/birthdefects/documents/MACDPcode0807.pdf*

Chen D, McMichael JC, VanDerMeid KR, *et al*. Evaluation of purified UspA from *Moraxella catarrhalis* as a vaccine in a murine model after active immunization. *Infect Immun* 1996;64:1900-05.

Daubin C, Parienti JJ, Vabret A, *et al*. Procalcitonin levels in acute exacerbation of COPD admitted in ICU: a prospective cohort study. *BMC Infect Dis* 2008;8:145.

Droemann D, Goldmann T, Tiedje T, *et al*. Toll-like receptor 2 expression is decreased on alveolar macrophages in cigarette smokers and COPD patients. *Resp Res* 2005;6:68-75.

EMA Guideline on the exposure to medicinal products during pregnancy: need for post-authorization data (Doc. Ref. EMEA/CHMP/313666/2005) 'adopted at Community level in May 2006);
http://www.ema.europa.eu/docs/en_GB/document_library/Regulatory_and_procedural_guideline/2009/11/WC500011303.pdf. Last accessed: 30-APR-2014.

Erb-Downward JR, Thompson DL, Han MK, *et al*. Analysis of the lung microbiome in the "healthy" smoker and in COPD. *PLoS One* 2011;6:e16384.

Forsgren A, Riesbeck K and Janson H. Protein D of *Haemophilus influenzae*: A Protective Nontypeable *H. influenzae* Antigen and a Carrier for Pneumococcal Conjugate Vaccines. *Clin Infect Dis* 2008;46:726-31.

Global Initiative for Chronic Obstructive Lung Disease (GOLD). From the Global Strategy for the diagnosis, management, and prevention of Chronic Obstructive Pulmonary Disease, updated 2014; *http://www.goldcopd.org/.* Last accessed: 30-APR-2014.

Hutchinson AF, Ghimire AK, Thompson MA, *et al*. A community-based, time-matched, case-control study of respiratory viruses and exacerbations of COPD. *Respir Med* 2007;101:2472-81.

Jurcisek JA, Bookwalter JE, Baker BD, *et al*. The PilA protein of non-typeable *Haemophilus influenzae* plays a role in biofilm formation, adherence to epithelial cells and colonization of the mammalian upper respiratory tract. *Mol Biol* 2007;65:1288-1299.

Ko FW, Ip M, Chan PK, *et al*. A 1-year prospective study of the infectious etiology in patients hospitalized with acute exacerbations of COPD. *Chest* 2007;131:44-52.

Larsen MV, Janner JH, Nielsen SD, *et al*. Bacteriology in acute exacerbation of chronic obstructive pulmonary disease in patients admitted to hospital. *Scand J Infect Dis* 2009;41:26-32.

Mannino DM, Homa DM, Akinbami LJ, *et al*. Chronic obstructive pulmonary disease surveillance-United States, 1971-2000. *MMWR Surveill Summ* 2002;51:1-16.

Miller MR, Hankinson J, Brusasco V, *et al.* Standardisation of spirometry. Series "ATS/ERS task force: standardisation of lung function testing" Editded by Brusasco V, Crapo R and Viegi G Number 2 in this Series. *Eur Respir J* 2005;26:319-338.

Moris P, van der Most R, Leroux-Roels I, *et al*. H5N1 influenza vaccine formulated with AS03 A induces strong cross-reactive and polyfunctional CD4 T-cell responses. *J Clin Immuno* 2011; 31: 443-54.

Murphy TF, Brauer AL, Grant BJ, *et al*. *Moraxella catarrhalis* in chronic obstructive pulmonary disease: burden of disease and immune response. *Am J Respir Crit Care Med* 2005;172:195-9.

Novotny LA, Jurcisek JA, Godfroid F, *et al.* Passive immunization with human anti-protein D antibodies induced by polysaccharide protein D conjugates protects chinchillas against otitis media after intranasal challenge with *Haemophilus influenzae. Vaccine* 2006;24:4804-11.

Novotny LA, Adams LD, Kang RD, *et al*. Epitope mapping immunodominant regions of the PilA protein of non-typeable *Haemophilus influenzae* (NTHi) to facilitate the design of two novel chimeric vaccine candidates. *Vaccine* 2009;28:279-289.

Nseir S, Ader F. Prevalence and outcome of severe chronic obstructive pulmonary disease exacerbations caused by multidrug-resistant bacteria. *Curr Opin Pulm Med* 2008;14:95-100.

Papi A, Bellettato CM, Braccioni F, *et al*. Infections and airway inflammation in chronic obstructive pulmonary disease severe exacerbations. *Am J Respir Crit Care Med* 2006;173:1114-21.

Rabe KF, Hurd S, Anzueto, *et al*. Global Strategy for the Diagnosis, Management, and Prevention of Chronic Obstructive Pulmonary Disease. GOLD Executive Summary. *Am J Respir Crit Care Med* 2007;176:532-555.

Ronander E, Brant M, Eriksson E, *et al.* non-typeable *Haemophilus influenzae* Adhesin Protein E: Characterization and Biological Activity. *J Infect Dis* 2009;199:522-31.

Rosell A, Monsó E, Soler N, *et al*. Microbiologic determinants of exacerbation in chronic obstructive pulmonary disease. *Arch Intern Med* 2005;165: 891-7.

Sapey E, Stockley RA. COPD exacerbations 2: aetiology. *Thorax* 2006;61:250-8.

Sethi S, Evans N, Grant BJ, *et al*. New strains of bacteria and exacerbations of chronic obstructive pulmonary disease. *N Engl J Med* 2002;347:465-71.

Sethi S, Murphy TF. Infection in the pathogenesis and course of chronic obstructive pulmonary disease. *N Engl J Med* 2008;359:2355-65.

Takanashi S, Hasegawa Y, Kanehira Y, *et al*. Interleukin-10 level in sputum is reduced in bronchial asthma, COPD and in smokers. *Eur Respir J* 1999;14:309-314.

Weinberger B, Herndler-Brandstetter D, Schwanninger A, *et al*. Biology of Immune Responses to Vaccines in Elderly Persons. *Clin Infect Dis* 2008;46:1078-84.

Wilkinson TM, Hurst JR, Perera WR, *et al*. Effect of interactions between lower airway bacterial and rhinoviral infection in exacerbations of COPD. *Chest* 2006;129:317-24.

APPENDIX A LABORATORY ASSAYS

Humoral immunity

Serological assays will be performed at GSK Biologicals' laboratory or in a GSK designated laboratory using assays as described in Table 10.

Anti-PD antibodies

Anti-PD antibodies will be determined using an ELISA assay developed by GSK Biologicals. Concentration of specific anti-PD antibodies will be determined, using in-house made reference serum. The current cut-off of the assay is ***153*** EL.U/mL. **(Amended 16 March 2016)**

Anti-PE antibodies

Anti-PE antibodies will be determined using an ELISA assay developed by GSK Biologicals. Concentration of specific anti-PE antibodies will be determined, using in-house made reference serum. The cut-off of the assay is 8 EL.U/mL.

Anti-PilA antibodies

Anti-PilA antibodies will be determined using an ELISA assay developed by GSK Biologicals. Concentration of specific anti-PilA antibodies will be determined, using an in-house made reference serum. The cut-off of the assay is 7 EL.U/mL.

Anti-UspA2 antibodies

Anti-UspA2 antibodies will be determined using an ELISA assay developed by GSK Biologicals. Concentration of specific anti-UspA2 antibodies will be determined, using an in-house made reference serum. The cut-off of the assay is 18 EL.U/mL.

Cell-mediated immunity

CMI assays will be performed at GSK Biologicals' laboratory or in a GSK designated laboratory using assays as described in Table 11.

The ICS staining assay will be used to assess CMI responses, using an adaptation of previously described methods [Moris, 2011]. After PBMC stimulation with the relevant antigens, the frequency of $CD4^+$ and/or $CD8^+$ T-cells expressing selected set of cytokines (such as IL-2, IL-13, IL-17, IFN-γ, TNF-α and CD40L) or selected combination of cytokines will be evaluated by flow cytometry.

APPENDIX B CLINICAL LABORATORIES (Amended 16 March 2016)

Table 24 GSK Biologicals' laboratories

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

Table 25 Outsourced laboratories

Laboratory	Address
Q² Solutions	Unit B1, Parkway West Industrial Estate Cranford Lane – Heston, Middlesex TW5 9QA United Kingdom

APPENDIX C FDA's Guidance for Industry: Toxicity Grading Scale for Healthy Adult and Adolescent Volunteers Enrolled in Preventive Vaccine Clinical Trials (September 2007)

This guidance represents the Food and Drug Administration's (FDA's) current thinking on this topic. It does not create or confer any rights for or on any person and does not operate to bind FDA or the public. You can use an alternative approach if the approach satisfies the requirements of the applicable statutes and regulations. If you want to discuss an alternative approach, contact the appropriate FDA staff. If you cannot identify the appropriate FDA staff, call the appropriate number listed on the title page of this guidance.

i. INTRODUCTION

Preventive vaccines are usually developed to prevent disease in a healthy population. The Office of Vaccines Research and Review, Center for Biologics Evaluation and Research, regulates preventive vaccines under authority of section 351 of the Public Health Service Act (42 U.S.C. 262), as well as specific sections of the Federal Food, Drug, and Cosmetic Act, and reviews investigational new drug applications (INDs) and biologics license applications (BLAs). (See, for example, Title 21 Code of Federal Regulations (CFR) Parts 312, 600, and 601). Most of the clinical trials of preventive vaccines conducted to support INDs and BLAs enroll healthy volunteers in all phases of vaccine testing. The enrollment of healthy volunteers warrants a very low tolerance for risk in those clinical trials.

This guidance provides you, sponsors, monitors, and investigators of vaccine trials, with recommendations on assessing the severity of clinical and laboratory abnormalities in healthy adult and adolescent volunteers enrolled in clinical trials. The grading system described in the table can also be useful in defining a particular study's stopping rules (e.g. a certain number of adverse events, as defined in the table, may call for stopping the study). Less extreme observations (e.g. mild) may not require discontinuing the study vaccine but can still contribute to evaluating safety by identifying parameters to focus upon in subsequent product development. Uniform criteria for categorizing toxicities in healthy volunteers can improve comparisons of safety data among groups within the same study and also between different studies. We, FDA, recommend using toxicity grading scale tables, provided below, as a guideline for selecting the assessment criteria to be used in a clinical trial of a preventive vaccine. We recommend incorporation of such appropriate, uniform, criteria into the investigational plan, case report forms, and CSRs and correspondence with FDA, sponsors, monitors, investigators, and Institutional Review Boards (IRBs).

This guidance finalizes the draft guidance of the same title dated April 2005 (70 FR 22664, May 2, 2005).

FDA's guidance documents, including this guidance, do not establish legally enforceable responsibilities. Instead, guidances describe FDA's current thinking on a topic and should be viewed only as recommendations, unless specific regulatory or statutory requirements

are cited. The use of the word should in FDA's guidances means that something is suggested or recommended, but not required.

ii. BACKGROUND

Standardised toxicity assessment scales have been widely used to evaluate products treating specific diseases. For example, the National Cancer Institute's Common Toxicity Criteria Scale and the Division of AIDS' Toxicity Grading Scale standardize the evaluation of adverse events among patients with cancer and HIV/AIDS, respectively (Refs. 1, 2). The defined toxicity parameters in those scales are designed for patients who may already experience mild, moderate, or severe adverse clinical or laboratory events due to the disease process, and may not be appropriate for healthy volunteers.

In the development of the toxicity grading scales for healthy volunteers, we chose parameter limit values based on published information, when such values were available (Refs. 1-6). For example, the Brighton Collaboration has developed case definitions and guidelines to evaluate some adverse events associated with administering vaccines (Ref. 3). In some cases, parameter limit values were based on clinical experience and experience reviewing vaccine clinical trials that enrol normal healthy subjects.

Toxicity grading scales for laboratory abnormalities should consider the local laboratory reference values when the parameter limit values are defined. The characterization of laboratory parameters among some populations of healthy adults and adolescents may require the exercise of clinical judgment, for example, consideration of the potential for ethnic differences in white blood cell (WBC) counts or gender differences in creatine phosphokinase (CPK) values.

iii. TOXICITY GRADING SCALE TABLES

Adverse events in a clinical trial of an investigational vaccine must be recorded and monitored and, when appropriate, reported to FDA and others involved in an investigation (sponsors, IRBs, and investigators). (See, for example, 21 CFR 312.32, 312.33, 312.50, 312.55, 312.56, 312.60, 312.62, 312.64, and 312.66). Although the use of a toxicity grading scale for adverse events would not replace these regulatory requirements, using a scale to categories adverse events observed during a clinical trial may assist you in monitoring safety and making required reports. Nonetheless, we believe that categorization or grading of data as outlined in this document is supplementary to and should not replace full and complete data analysis.

These guidelines for toxicity grading scales are primarily intended for healthy adult and adolescent volunteers. The parameters in the tables below are not necessarily applicable to every clinical trial of healthy volunteers. The parameters monitored should be appropriate for the specific study vaccine. For some preventive vaccines under development, it may be appropriate to include additional parameters to be monitored during a clinical trial or to alter the choice of values in the toxicity table. For example, additional parameters might be added based on one or more of the following: safety signals observed in pre-clinical toxicology studies, the biological plausibility of the occurrence of certain adverse events, or previous experience with a similar licensed product.

As discussed above, the tables do not represent a recommendation to monitor all the listed parameters in all clinical trials of healthy volunteers, nor do the tables represent all possible parameters to be monitored. In addition, these tables do not represent study inclusion or exclusion criteria. We recommend that the parameters monitored be appropriate for the study vaccine administered to healthy volunteers participating in the clinical trial.

Tables for Clinical Abnormalities

Note from the sponsor: The tables in this section of the guidance will not be used in this particular study. Instead, the parameters as provided in the study protocol are to be used.

Tables for Laboratory Abnormalities

The laboratory values provided in the tables below serve as guidelines and are dependent upon institutional normal parameters. Institutional normal reference ranges should be provided to demonstrate that they are appropriate.

Table 26 FDA toxicity grading scales for haematology/ biochemistry parameters

Serum *	Mild (Grade 1)	Moderate (Grade 2)	Severe (Grade 3)	Potentially Life Threatening (Grade 4)**
Sodium – Hyponatremia mEq/L	132 – 134	130 – 131	125 – 129	< 125
Sodium – Hypernatremia mEq/L	144 – 145	146 – 147	148 – 150	> 150
Potassium – Hyperkalemia mEq/L	5.1 – 5.2	5.3 – 5.4	5.5 – 5.6	> 5.6
Potassium – Hypokalemia mEq/L	3.5 – 3.6	3.3 – 3.4	3.1 – 3.2	< 3.1
Glucose – Hypoglycemia mg/dL	65 – 69	55 – 64	45 – 54	< 45
Glucose – Hyperglycemia Fasting – mg/dL Random – mg/dL	100 – 110 110 – 125	111 – 125 126 – 200	>125 >200	Insulin requirements or hyperosmolar coma
Blood Urea Nitrogen BUN mg/dL	23 – 26	27 – 31	> 31	Requires dialysis
Creatinine – mg/dL	1.5 – 1.7	1.8 – 2.0	2.1 – 2.5	> 2.5 or requires dialysis
Calcium – hypocalcemia mg/dL	8.0 – 8.4	7.5 – 7.9	7.0 – 7.4	< 7.0
Calcium – hypercalcemia mg/dL	10.5 – 11.0	11.1 – 11.5	11.6 – 12.0	> 12.0
Magnesium – hypomagnesemia mg/dL	1.3 – 1.5	1.1 – 1.2	0.9 – 1.0	< 0.9
Phosphorous – hypophosphatemia mg/dL	2.3 – 2.5	2.0 – 2.2	1.6 – 1.9	< 1.6
CPK – mg/dL	1.25 – 1.5 x ULN	1.6 – 3.0 x ULN	3.1 –10 x ULN	> 10 x ULN
Albumin – Hypoalbuminemia g/dL	2.8 – 3.1	2.5 – 2.7	< 2.5	--
Total Protein – Hypoproteinemia g/dL	5.5 – 6.0	5.0 – 5.4	< 5.0	--
Alkaline phosphate – increase by factor	1.1 – 2.0 x ULN	2.1 – 3.0 x ULN	3.1 – 10 x ULN	> 10 x ULN
Liver Function Tests –ALT, AST increase by factor	1.1 – 2.5 x ULN	2.6 – 5.0 x ULN	5.1 – 10 x ULN	> 10 x ULN
Bilirubin – when accompanied by any increase in Liver Function Test increase by factor	1.1 – 1.25 x ULN	1.26 – 1.5 x ULN	1.51 – 1.75 x ULN	> 1.75 x ULN
Bilirubin – when Liver Function Test is normal; increase by factor	1.1 – 1.5 x ULN	1.6 – 2.0 x ULN	2.0 – 3.0 x ULN	> 3.0 x ULN
Cholesterol	201 – 210	211 – 225	> 226	---
Pancreatic enzymes – amylase, lipase	1.1 – 1.5 x ULN	1.6 – 2.0 x ULN	2.1 – 5.0 x ULN	> 5.0 x ULN

* The laboratory values provided in the tables serve as guidelines and are dependent upon institutional normal parameters. Institutional normal reference ranges should be provided to demonstrate that they are appropriate.
** The clinical signs or symptoms associated with laboratory abnormalities might result in characterisation of the laboratory abnormalities as Potentially Life Threatening (Grade 4). For example, a low sodium value that falls within a grade 3 parameter (125-129 mE/L) should be recorded as a grade 4 hyponatremia event if the subject had a new seizure associated with the low sodium value.
ULN is the upper limit of the normal range.

Haematology *	Mild (Grade 1)	Moderate (Grade 2)	Severe (Grade 3)	Potentially Life Threatening (Grade 4)
Haemoglobin (Female) - gm/dL	11.0 – 12.0	9.5 – 10.9	8.0 – 9.4	< 8.0
Haemoglobin (Female) change from baseline value - gm/dL	Any decrease – 1.5	1.6 – 2.0	2.1 – 5.0	> 5.0
Haemoglobin (Male) - gm/dL	12.5 – 13.5	10.5 – 12.4	8.5 – 10.4	< 8.5
Haemoglobin (Male) change from baseline value – gm/dL	Any decrease – 1.5	1.6 – 2.0	2.1 – 5.0	> 5.0
WBC Increase - cell/mm^3	10,800 – 15,000	15,001 – 20,000	20,001 – 25, 000	> 25,000
WBC Decrease - cell/mm^3	2,500 – 3,500	1,500 – 2,499	1,000 – 1,499	< 1,000
Lymphocytes Decrease - cell/mm^3	750 – 1,000	500 – 749	250 – 499	< 250
Neutrophils Decrease - cell/mm^3	1,500 – 2,000	1,000 – 1,499	500 – 999	< 500
Eosinophils - cell/mm^3	650 – 1500	1501 - 5000	> 5000	Hypereosinophilic
Platelets Decreased - cell/mm^3	125,000 – 140,000	100,000 – 124,000	25,000 – 99,000	< 25,000
PT – increase by factor (prothrombin time)	1.0 – 1.10 x ULN	1.11 – 1.20 x ULN	1.21 – 1.25 x ULN	> 1.25 ULN
PTT – increase by factor (partial thromboplastin time)	1.0 – 1.2 x ULN	1.21 – 1.4 x ULN	1.41 – 1.5 x ULN	> 1.5 x ULN
Fibrinogen increase - mg/dL	400 – 500	501 – 600	> 600	--
Fibrinogen decrease - mg/dL	150 – 200	125 – 149	100 – 124	< 100 or associated with gross bleeding or disseminated intravascular coagulation (DIC)

* The laboratory values provided in the tables serve as guidelines and are dependent upon institutional normal parameters. Institutional normal reference ranges should be provided to demonstrate that they are appropriate. ULN is the upper limit of the normal range.

iv. REFERENCES for the Appendix C

1. National Cancer Institute Common Toxicity Criteria, April 30, 1999. (*http://ctep.cancer.gov/reporting/CTC-3.html*).
2. Division of AIDS Table for Grading Severity of Adult Adverse Experiences; August 1992. (*http://rcc.tech-res-intl.com/tox_tables.htm*).
3. The Brighton Collaboration. Finalized Case Definitions and Guidelines. (*http://brightoncollaboration.org/internet/en/index/definition_guidelines.html*).
4. HIV Vaccine Trials Network Table for Grading Severity of Adverse Experiences; September 18, 2002. (*http://rcc.tech-res-intl.com/tox_tables.htm*).
5. Division of AIDS Table for Grading the Severity of Adult and Pediatric Adverse Events, December 2004. (*http://www3.niaid.nih.gov/research/resources/DAIDSClinRsrch/PDF/Safety/DAIDSAEGradingTable.pdf*).
6. Kratz A, Ferraro M, Sluss PM, Lewandrowski KB. Laboratory Reference Values. *Engl J Med* 2004;351:1548-1563.

APPENDIX D AMENDMENTS AND ADMINISTRATIVE CHANGES TO THE PROTOCOL

GlaxoSmithKline Biologicals Clinical Research & Development **Protocol Amendment 1**	
eTrack study number and Abbreviated Title(s)	201281 (NTHI MCAT-001)
Amendment number:	Amendment 1
Amendment date:	30 July 2015
Co-ordinating author:	PPD, Scientific Writer, XPE Pharma for GSK Biologicals

Rationale/background for changes:

- The definition of adequate contraception has been aligned with the Heads of Medicines Agencies (HMA) Clinical Trial Facilitation Group (CTFG) guidance 'Recommendations related to contraception and pregnancy testing in clinical trials' (2014).
- The blood sample volume has been adapted as follows:
  - Safety samples: ~5.5 mL per visit instead of ~10 mL.
  - CMI sample: ~20 mL per visit instead of ~30 mL.

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

LIST OF ABBREVIATIONS

ATS	***American Thoracic Society***
ERS	***European Respiratory Society***

GLOSSARY OF TERMS

Adequate contraception

Adequate contraception is defined as:

1. a contraceptive method with failure rate of less than 1% per year when used consistently and correctly and when applicable, in accordance with the product label for example:

- abstinence from penile-vaginal intercourse, when this is their preferred and usual lifestyle,
- oral contraceptives, either combined or progestogen alone,
- injectable progestogen,
- implants of etenogestrel or levonorgestrel,
- estrogenic vaginal ring,
- percutaneous contraceptive patches,
- intrauterine device or intrauterine system,
- ***current tubal ligation,***
- male partner sterilisation prior to the female subject's entry into the study, and this male is the sole partner for that subject,

  The information on the male sterility can come from the site personnel's review of the subject's medical records; or interview with the subject on her medical history.

2. a contraceptive method with failure rate of more than 1% per year but still considered as acceptable birth control method:

- male condom combined with a vaginal spermicide (foam, gel, film, cream or suppository),
- male condom combined with a female diaphragm, either with or without a vaginal spermicide (foam, gel, film, cream, or suppository).

Adequate contraception does not apply to subjects of child bearing potential with same sex partners, when this is their preferred and usual lifestyle.

Section 4.2.1 Healthy adults 18-40 years old (Step 1)

- Female subjects of non-childbearing potential may be enrolled in the study.
  - Non-childbearing potential is defined as pre-menarche, ~~current tubal ligation,~~ hysterectomy, ovariectomy or post-menopause.

Section 4.2.2 Current and former smokers 50-70 years old (Step 2)

- Female subjects of non-childbearing potential may be enrolled in the study.
  - Non-childbearing potential is defined as pre-menarche, hysterectomy, ~~current tubal ligation,~~ ovariectomy or post-menopause.

Section 5.5 Outline of study procedures

Table 5 List of study procedures for healthy adults 18-40 years old (Step 1)

Epoch	*Epoch 001*							*Epoch 002*	
Type of contact	Screening Visit	Visit 1	Visit 2	Visit 3	Visit 4	Visit 5	Visit 6	Visit 7	Visit 8
Time points	pre-Day 0	Day 0	Day 7	Day 30	Day 60	Day 67	Day 90	Day 210	Day 420
Sampling time points	Screening	Pre	Post-Vacc I			Post-Vacc II			
Informed consent	●								
Check inclusion/exclusion criteria	●[(d)]	O							
Collect demographic data	●								
Medical history	O[(d)]	●							
Measure/record height and weight	●								●
Physical examination	●[(d, e)]	O	O	O	O	O	O	O	O
Urine pregnancy test [(a)]	●	●			●				
Check contraindications		O			O				
Pre-vaccination body temperature		●			●				
Treatment number allocation [(b)]		●			●				
Blood sampling for safety assessment (~~10~~***5.5*** mL)	●[(d)]	●	●		●	●		●	●
Blood sampling for antibody determination and assay development (~20 mL)		●		●	●		●	●	●
Vaccine administration		●			●				
Distribution of Diary Cards [(c)]		O	O		O	O			
Return of Diary Cards			O	O		O	O		
Diary Card transcription by investigator			●	●		●	●		
Recording of AEs		●	●	●	●	●	●		
Recording of SAEs	●[(f)]	●	●	●	●	●	●	●	●
Recording of pIMDs		●	●	●	●	●	●	●	●
Recording of pregnancies		●	●	●	●	●	●	●	●
Record concomitant medication/vaccination [(g)]		●	●	●	●	●	●	●	●
Record intercurrent medical conditions requiring medical attention		●	●	●	●	●	●	●	●
Screening conclusion	●								
Study conclusion									●

Note: The double-line borders following Day 90 and Day 420 indicates the statistical analyses which will be performed on all data (i.e. data that are as clean as possible) obtained up to Day 90 and Day 420, respectively.
Vacc: Vaccination; **AEs:** Adverse events; **SAEs:** Serious adverse events; **pIMDs:** potential immune-mediated disease.
● 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.

[a] A urine pregnancy test will be performed only for women of childbearing potential.
[b] Treatment number allocation with randomisation at Visit 1 (Day 0); treatment number allocation without randomisation at Visit 4 (Day 60).
[c] Distribution of **2** Diary Cards after each vaccination:

- a 1st one distributed on the day of vaccination for recording of solicited and unsolicited AEs and concomitant medications/ products and vaccinations from Day 0 to Day 6 after each vaccination.
- a 2nd one distributed on the 7 day post-vaccination visit for recording of unsolicited AEs and concomitant medications/ products and vaccinations from Day 7 to Day 29 after each vaccination.

[d] Screening evaluations may be completed 1 to 28 days before Day 0. Site staff should allow sufficient time between the Screening and Visit 1 to receive and review screening safety laboratory test results. If a delay occurs such that the interval between Screening and the Visit 1 vaccination exceeds 28 days, a re-screening visit should be scheduled before Visit 1 during which blood sample collection for safety laboratory assessment must be repeated; an interim medical history and physical examination must be obtained and inclusion / exclusion criteria must be re-reviewed.
[e] Complete physical examination including vital signs.
[f] From Screening to Visit 1, only those SAEs that are considered related to study participation or to concomitant use of GSK products or any fatal events need to be recorded. GSK products or any fatal event need to be recorded.
[g] Concomitant medication products/vaccination as indicated in Section 6.6.1 need to be recorded.

Table 6 List of study procedures for current or former smokers 50-70 years old (Step 2)

Epoch	*Epoch 001*							*Epoch 002*	
Visit 6	**Screening Visit**	**Visit 1**	**Visit 2**	**Visit 3**	**Visit 4**	**Visit 5**	**Visit 6**	**Visit 7**	**Visit 8**
Time points	**pre-Day 0**	**Day 0**	**Day 7**	**Day 30**	**Day 60**	**Day 67**	**Day 90**	**Day 210**	**Day 420**
Sampling time points	**Screening**	**Pre**	**Post-Vacc I**			**Post-Vacc II**			
Informed consent	●								
Check inclusion/exclusion criteria	●[e]	O							
Collect demographic data	●								
Medical history	O[e]	●							
Smoking status	●							●	●
Smoking exposure history	●								
Measure/record height and weight	●								●
Spirometry	●								●
Physical examination	●[e, f]	O	O	O	O	O	O	O	O
Urine pregnancy test [a]	●	●			●				
Check contraindications		O			O				
Pre-vaccination body temperature		●			●				
Treatment number allocation[b]		●			●				
Blood sampling for safety assessment (~~10~~***5.5*** mL)	●[e]	●	●		●	●		●	●
Blood sampling for antibody determination and assay development (~20 mL)		●		●	●		●	●	●
Blood sampling for CMI response (~~30~~***20*** mL)[c]		●			●		●	●	●
Vaccine administration		●			●				
Distribution of Diary Cards[d]		O	O		O	O			
Return of Diary Cards			O	O		O	O		
Diary Card transcription by investigator			●	●		●	●		
Recording of AEs		●	●	●	●	●	●		
Recording of SAEs	●[g]	●	●	●	●	●	●	●	●
Recording of pIMDs		●	●	●	●	●	●	●	●
Recording of pregnancies		●	●	●	●	●	●	●	●
Record concomitant medication/vaccination[h]		●	●	●	●	●	●	●	●
Record intercurrent medical conditions requiring medical attention		●	●	●	●	●	●	●	●

Epoch	*Epoch 001*							*Epoch 002*	
Visit 6	**Screening Visit**	**Visit 1**	**Visit 2**	**Visit 3**	**Visit 4**	**Visit 5**	**Visit 6**	**Visit 7**	**Visit 8**
Time points	**pre-Day 0**	**Day 0**	**Day 7**	**Day 30**	**Day 60**	**Day 67**	**Day 90**	**Day 210**	**Day 420**
Sampling time points	**Screening**	**Pre**	**Post-Vacc I**			**Post-Vacc II**			
Screening conclusion	●								
Study conclusion									●

Note: The double-line borders following Day 90 and Day 420 indicates the statistical analyses which will be performed on all data (i.e. data that are as clean as possible) obtained up to Day 90 and Day 420, respectively.
Vacc: Vaccination; **CMI:** Cell-mediated immunity; **AEs:** Adverse events; **SAEs:** Serious adverse events; **pIMDs:** potential immune-mediated disease.
● 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.
[(a)] A urine pregnancy test will be performed only for women of childbearing potential.
[(b)] Treatment number allocation with randomisation at Visit 1 (Day 0); treatment number allocation without randomisation at Visit 4 (Day 60).
[(c)] Only for subjects belonging to the CMI sub-cohort.
[(d)] Distribution of **2** Diary Cards after each vaccination:

- a 1st one distributed on the day of vaccination for recording of solicited and unsolicited AEs and concomitant medications/ products and vaccinations from Day 0 to Day 6 after each vaccination.
- a 2nd one distributed on the 7 day post-vaccination visit for recording of unsolicited AEs and concomitant medications/ products and vaccinations from Day 7 to Day 29 after each vaccination.

[(e)] Screening evaluations may be completed 1 to 28 days before Day 0. Site staff should allow sufficient time between the Screening and Visit 1 to receive and review screening safety laboratory test results. If a delay occurs such that the interval between Screening and the Visit 1 vaccination exceeds 28 days, a re-screening visit should be scheduled before Visit 1 during which blood sample collection for safety laboratory assessment must be repeated; an interim medical history and physical examination must be obtained and inclusion / exclusion criteria must be re-reviewed.
[(f)] Complete physical examination including vital signs.
[(g)] From Screening to Visit 1, only those SAEs that are considered related to study participation or to concomitant use of GSK products or any fatal events need to be recorded. GSK products or any fatal event need to be recorded.
[(h)] Concomitant medication products/vaccination as indicated in Section 6.6.1 need to be recorded.

Section 5.6.8 Spirometry

For all subjects in Step 2, spirometry to assess FEV_1 and FVC will be performed using techniques that meet ***American Thoracic Society (ATS)/European Respiratory Society (ERS)*** published standards [Miller, 2005].

Section 5.6.14.1 Blood sampling for safety assessment

A volume of approximately ***5.5***~~10~~ mL of whole blood should be drawn at each pre-defined time point.

5.6.14.3 Blood sampling for CMI

A volume of approximately ***20***~~30~~ mL of whole blood should be drawn from all subjects included in the immunogenicity sub-cohort for CMI at each pre-defined time point. The blood should be stored at the investigator's site at room temperature and it must not be centrifuged. Samples will be shipped at room temperature (20 to 25°C) to the designated laboratory for cell and plasma separation to be performed within 24 hours of collection.

Section 5.7.2 Biological samples

Table 8 Biological samples

Sample type	Quantity	Unit	Time point	Sub-cohort
Blood for safety assessment	~***5.5***~~10~~	mL	Screening Visit (pre-Day 0)	All screened subjects
			Visit 1 (Day 0)	All enrolled subjects
			Visit 2 (Day 7)	
			Visit 4 (Day 60)	
			Visit 5 (Day 67)	
			Visit 7 (Day 210)	
			Visit 8 (Day 420)	
Blood for antibody determination and assay development	~20	mL	Visit 1 (Day 0))	All enrolled subjects
			Visit 3 (Day 30)	
			Visit 4 (Day 60)	
			Visit 6 (Day 90)	
			Visit 7 (Day 210)	
			Visit 8 (Day 420)	
Blood for CMI response	~***20***~~30~~	mL	Visit 1 (Day 0))	Sub-cohort for CMI*
			Visit 4 (Day 60)	
			Visit 6 (Day 90)	
			Visit 7 (Day 210)	
			Visit 8 (Day 420)	

* Refer to Section 4.1 for sub-cohort description.

GlaxoSmithKline Biologicals

Clinical Research & Development

Protocol Amendment 2

eTrack study number and Abbreviated Title(s)	201281 (NTHI MCAT-001)
Amendment number:	Amendment 2
Amendment date:	16 March 2016
Co-ordinating author:	PPD, Scientific Writer, XPE Pharma for GSK Biologicals

Rationale/background for changes:

- The spirometry test must be of good quality. If this is not the case, a re-test should be done. This re-test is covered by the current Informed Consent Form.
- In compliance with ICH requirements, the protocol mentions that all results will be presented in an integrated report at the end of the study.
- A description of how subjects will be included in the CMI sub-cohort and a description of how the sub-cohort will represent the fully randomized study groups has been added.
- Following re-development and re-validation of the anti-PD ELISA, a new cut-off was defined.
- Additional administrative changes have been performed:
  - The IND number has been added
  - The list of contributing authors as well as Sponsor signatory has been updated
  - GSK Biologicals' "Global Vaccines Clinical Laboratories (GVCL)" has been renamed as "Clinical Laboratory Sciences (CLS)"
  - Name "Quest Diagnostic Limited" has been replaced by "Q² Solutions" due to its merge with QLab
  - Reference to the GSK Biologicals' Laval laboratory was removed, as this laboratory will not be used in the study. In addition, this laboratory is no longer part of GSK Biologicals' laboratories.

    In addition, minor edits in other sections were made for clarification.

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

Cover Page, Protocol Amendment 2 Sponsor Signatory Approval and Protocol Amendment 2 Investigator Agreement

Investigational New Drug (IND) number	***16531***

Contributing authors

Contributing authors (continued)

- PPD, Director Clinical Research & Translational Science
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- PPD, Study Delivery Lead
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- PPD, ~~GVCL~~ ***CLS Clinical Readout Team Leader*** ~~Project Manager~~
- PPD, ~~GVCL~~ ***CLS Laboratory*** Study Manager

Protocol Amendment 2 Sponsor Signatory Approval

Sponsor signatory	~~Jeanne-Marie Devaster, Director Clinical Research and Translational Science~~ ***Ashwani Kumar Arora, Clinical and Epidemiology Project Lead***

List of Abbreviations

CLS	***Clinical Laboratory Sciences***
~~GVCL~~	~~Global Vaccine Clinical Laboratories~~

Section 4.1 Number of subjects/centres

Approximately 50% of the subjects in each group of Step 2 will be part of a **sub-cohort for CMI analysis**. ***Subjects will be selected based on the enrolment order, i.e. the first 50% of the participants (approximately the first 15 subjects/group) will be enrolled in the sub-cohort for CMI.*** An additional blood sample for CMI analysis will be taken from

those subjects at each of the CMI blood sampling time points (i.e. Day 0, Day 60, Day 90, Day 210 and Day 420).

Section 5.2.2.2.1 Study group and treatment number allocation

As several minimisation factors have been defined for the treatment allocation at Step 2, it was decided at the design phase not to minimise for the CMI group allocation. This wouldn't be feasible from an implementation point of view.

Section 5.5 Outline of study procedures

Table 6 List of study procedures for current or former smokers 50-70 years old (Step 2)

Epoch	*Epoch 001*							*Epoch 002*	
Visit 6	**Screening Visit**	**Visit 1**	**Visit 2**	**Visit 3**	**Visit 4**	**Visit 5**	**Visit 6**	**Visit 7**	**Visit 8**
Time points	**pre-Day 0**	**Day 0**	**Day 7**	**Day 30**	**Day 60**	**Day 67**	**Day 90**	**Day 210**	**Day 420**
Sampling time points	**Screening**	**Pre**	**Post-Vacc I**			**Post-Vacc II**			
Informed consent	●								
Check inclusion/exclusion criteria	●(e)	O							
Collect demographic data	●								
Medical history	O(e)	●							
Smoking status	●							●	●
Smoking exposure history	●								
Measure/record height and weight	●								●
Spirometry *(i)*	●								●

(i) This test should be repeated on another day if the first attempt doesn't meet quality requirements as described by ATS/ERS standards (Miller, 2005).

Section 5.6.8 Spirometry

The spirometry test must be of good quality. If this is not the case, a re-test should be done.

Section 5.7.1 Use of specific study materials

When materials are provided by GSK Biologicals ***or by the Central laboratory***, it is MANDATORY that all clinical samples (including serum samples) be collected and stored exclusively using those materials in the appropriate manner.

Section 5.7.3 Laboratory assays

Table 10 Humoral Immunity (antibody determination)

System	Component	Method	Kit / Manufacturer	Unit	Cut-off	Laboratory*
Serum	Anti-PD	ELISA	In house	EL.U/mL	***153*** ~~100***~~	GSK Biologicals**
Serum	Anti-PE	ELISA	In house	EL.U/mL	8	GSK Biologicals**
Serum	Anti-PilA	ELISA	In house	EL.U/mL	7	GSK Biologicals**
Serum	Anti-UspA2	ELISA	In house	EL.U/mL	18	GSK Biologicals**

ELISA: Enzyme-Linked Immunosorbent Assay; EL.U/mL = ELISA unit per millilitre;
* Refer to APPENDIX B for the laboratory addresses.
** GSK Biologicals laboratory refers to the ***Clinical Laboratory Sciences (CLS) Laboratories*** ~~Global Vaccines Clinical Laboratories (GVCL)~~ in Rixensart, Belgium; Wavre, Belgium.; ~~Laval, Canada~~ or a designated laboratory.
~~*** A new cut-off may be defined after anti-PD-ELISA assay re-development.~~

Table 11 Cell-Mediated Immunity (CMI)

System	Component	Scale	Method	Unit	Laboratory*
PBMC	Specific $CD4^+/CD8^+$ T-cells	Quantitative	Flow cytometry	Number of specific $CD4^+/CD8^+$ T cells /10^6	GSK Biologicals**

PBMC = peripheral blood mononuclear cell.
* Refer to APPENDIX B for the laboratory addresses.
** GSK Biologicals laboratory refers to the ***Clinical Laboratory Sciences (CLS) Laboratories*** ~~Global Vaccines Clinical Laboratories (GVCL)~~ in Rixensart, Belgium; Wavre, Belgium~~; Laval, Canada~~ or a designated laboratory.

Section 10.6.3.1 Humoral immune response

The difference in terms of GMCs will be evaluated, one month post-Dose 2, by computing the 95% CIs of the GMC ratio between groups for Step 2 by using a one-way ANCOVA model on the logarithm10 transformation of the concentrations/ titres. The ANCOVA model will include the group category, the age category, the smoking status and FEV_1/FVC (≥ 0.7 or < 0.7) ***and the pre-Dose 1 concentration (as covariate)*** as fixed effects ~~and the pre-Dose 1 concentration as regressor~~. The groups will be considered significantly different if the 95% CI for the GMC ratio between the two groups does not contain the value 1.

Section 10.8.1 Sequence of analysis

All study results will be presented in an integrated study report at the end of the study.

Appendix A Laboratory Assays

Humoral immunity

Anti-PD antibodies

Anti-PD antibodies will be determined using an ELISA assay developed by GSK Biologicals. Concentration of specific anti-PD antibodies will be determined, using in-house made reference serum. The current cut-off of the assay is ***153*** ~~100~~ EL.U/mL.

~~This assay will however be re-worked according to the latest validation standards and a new cut-off might be defined based on a precise and accurate limit of quantification.~~

Appendix B Clinical laboratories

Table 24 GSK Biologicals' laboratories

Laboratory	Address
GSK Biologicals Global Vaccine Clinical Laboratory, Rixensart	Biospecimen Reception - B7/44 Rue de l'Institut, 89 - B-1330 Rixensart - Belgium
~~GSK Biologicals Global Vaccine Clinical Laboratory, North America - Laval~~	~~Biospecimen Reception - Clinical Serology 525 Cartier blvd West - Laval - Quebec - Canada - H7V 3S8~~

Table 25 Outsourced laboratories

Laboratory	Address
~~Quest Diagnostic Limited~~ ***Q² Solutions***	Unit B1, Parkway West Industrial Estate Cranford Lane – Heston, Middlesex TW5 9QA United Kingdom

References

~~Heads of Medicines Agencies (HMA) Clinical Trial Facilitation Group (CTFG) guidance - Recommendations related to contraception and pregnancy testing in clinical trials. (2014). http://www.hma.eu/fileadmin/dateien/Human_Medicines/01-About_HMA/Working_Groups/CTFG/2014_09_HMA_CTFG_Contraception.pdf~~

Protocol Amendment 2 Sponsor Signatory Approval

eTrack study number and Abbreviated Title	201281 (NTHI MCAT-001)
IND number	***16531***
EudraCT number	2015-000378-36
Date of protocol amendment	Amendment 2 Final 16 March 2016
Detailed Title	A Phase I, randomised, observer-blind, placebo-controlled, multi-centre study to evaluate the safety, reactogenicity and immunogenicity of GSK Biologicals' GSK3277511A investigational vaccine when administered intramuscularly according to a 0, 2 months schedule in adults.
Sponsor signatory (Amended 16 March 2016)	***Ashwani Kumar Arora, Clinical and Epidemiology Project Lead***
Signature	PPD
Date	18 March 2016

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