CLINICAL STUDY PROTOCOL

Primary Study Intervention(s)	Salbutamol administered via metered dose inhalers containing propellant HFA-152a (test).
Other Study Intervention(s)	Salbutamol administered via metered dose inhalers containing propellant HFA-134a (reference).
Study Identifier	219728
EU CT Number	2023-508279-36-00
Approval Date	02 May 2024
Title	A Phase 1, randomized, open-label, single dose, 2-treatment arm (200 µg and 800 µg), 4-way cross-over study in healthy participants aged 18 to 55 to compare the pharmacokinetics of salbutamol administered via metered dose inhalers containing propellants HFA-152a (test) and HFA-134a (reference).
Compound Number/Name	AH3365
Brief Title	A study to compare the pharmacokinetics of salbutamol administered via metered dose inhalers containing propellants HFA-152a (test) or HFA-134a (reference) in healthy participants aged 18 to 55 inclusive.
Sponsor	GSK Research & Development Limited 980 Great West Road, Brentford, Middlesex, TW8 9GS, UK
Sponsor signatory	Oscar Della Pasqua, MD, PhD Executive Director Clinical Pharmacology Modeling & Simulation

Medical monitor name and contact can be found in local study contact information document.

Protocol Amendment 1 Investigator Agreement

I agree:

- To conduct the study in compliance with this protocol, any future protocol amendments, with the terms of the clinical trial agreement and with any other study conduct procedures and/or study conduct documents provided by GSK.
- To assume responsibility for the proper conduct of the study at this site.
- That I am aware of and will comply with GCP and all applicable regulatory requirements.
- That I will comply with the terms of the site agreement.
- To comply with local bio-safety legislation.
- To ensure that all persons assisting me with the study are adequately informed about the GSK study intervention and other study-related duties and functions as described in the protocol.
- To supervise any individual or party to whom I have delegated study-related duties and functions conducted at the study site.
- To ensure that any individual or party to whom I have delegated study-related duties and functions conducted at the study site are qualified to perform those study-related duties and functions.
- 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 on-site or elsewhere without the approval of GSK and the express physical or digital informed consent of the participant.
- To perform no biological assays on the clinical samples other than those described in the protocol or its amendment(s).
- To co-operate with representative(s) of GSK in the monitoring and data management processes of the study with respect to data entry and resolution of queries about the data.
- To have control of all essential documents and records generated under my responsibility before, during, and after the study.
- 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 study intervention(s), and more generally about their financial ties with the sponsor. GSK will use and disclose the information solely for the purpose of complying with regulatory requirements.

Hence, I:

- Agree to supply GSK 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 study and for 1 year following completion of the study.
- Agree that GSK may disclose any information about such ownership interests and financial ties to regulatory authorities.
- Agree to provide GSK with an updated Curriculum Vitae and all other documents required by regulatory agencies for this study.

Study identifier	219728
EU CT number	2023-508279-36-00
Approval date	02 May 2024
Title	A Phase 1, randomized, open-label, single dose, 2-treatment arm (200 µg and 800 µg), 4-way cross-over study in healthy participants aged 18 to 55 to compare the pharmacokinetics of salbutamol administered via metered dose inhalers containing propellants HFA-152a and HFA-134a (reference).
Investigator name	
Signature	
Date of signature (DD Month YYYY)	

Protocol Amendment Summary Of Changes Table

DOCUMENT HISTORY	
Document	**Date of Issue**
Amendment 1	02 May 2024
Original Protocol	18 January 2024

Type of Protocol Amendment	Numbering	Type of changes
Global	Amendment 1	New changes for all

Amendment 1 (02 May 2024)

Overall rationale for the current Amendment:

The objective of this amendment is to align the time period for collecting pregnancy information for female participants with the time period for post-intervention contraception. In addition, parameter specifications for microscopic evaluation have been removed.

LIST OF MAIN CHANGES IN THE PROTOCOL AND THEIR RATIONALE:

Section # and title	Description of change	Brief rationale
8.4.6 Pregnancy	Changed time period to collect pregnancy information from 90 days to 48 hours.	To align with the time period of post-intervention contraception.
10.2 Clinical laboratory test	Removal of parameter specifications for microscopic evaluation.	Specific parameters will be determined in accordance with clinical laboratory procedures.
11. References	References update.	Administrative update.
4.3 Justification for dose and 8.5 Pharmacokinetics	Other typographical changes.	Correction of units.

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LIST OF ABBREVIATIONS AND DEFINITIONS OF TERMS

Abbreviation	Definition
ADE	Adverse device effect
AE	Adverse event
AESI	Adverse event of special interest
ALT	Alanine transaminase
AM	Arithmetic mean
ATS	American Thoracic Society
AUC(0-last)	Area under the curve (from zero to last)
AUC(0-∞)	Area under the curve (from zero to infinity)
AxMP	Auxiliary medicinal product
BMI	Body mass index
Cmax	Maximum plasma concentration
CA	Competent authority
CFC	Chlorofluorocarbon
CFR	Code of Federal Regulations
CI	Confidence intervals
CIOMS	Council for International Organizations of Medical Sciences
CONSORT	Consolidated Standards of Reporting Trials
COVID-19	Coronavirus disease 2019
CPK	Creatine phosphokinase
CRF	Case report form
CSR	Clinical study report
CVw	Within-subject coefficient of variation
eCRF	Electronic case report form
ECG	Electrocardiogram
EMA	European Medicines Agency
ERS	European Respiratory Society
FDA	Food and Drug Administration, United States of America
FSFV	First subject first visit
FSH	Follicle stimulating hormone
GCP	Good Clinical Practice
GM	Geometric mean
GMR	Geometric mean ratio
GWP	Global warming potential
HBsAg	Hepatitis B surface antigen

Abbreviation	Definition
hCG	Human Chorionic Gonadotropin
HCV	Hepatitis C virus
HFA	Hydrofluoroalkane
HFA-134a	1,1,1,2-tetrafluoroethane
HFA-152a	1,1-difluoroethane
HIV	Human immunodeficiency virus
HR	Heart rate
HRT	Hormonal Replacement Therapy
ICE	Intercurrent event
ICF	Informed consent form
ICH	International Council for Harmonization
ICMJE	International Committee of Medical Journal Editors
ICSR	Individual case safety reports
IDSL	Integrated Data Standards Library
IEC	Independent Ethics Committee
IFU	Instructions for use
IgM	Immunoglobulin M
IMP	Investigational Medicinal Product
INR	International Normalized Ratio
IRB	Institutional Review Board
LDH	Lactate Dehydrogenase
LLOQ	Lower Limit of Quantification
LSLV	Last Subject Last Visit
MDI	Metered Dose Inhaler
MHR	Maximum Heart Rate
MedDRA	Medical Dictionary for Regulatory Activities
MSDS	Material Safety Data Sheet
NIMP	Non-investigational medicinal product
PCR	Polymerase Chain Reaction
PD	Pharmacodynamic(s)
PK	Pharmacokinetic(s)
PR	Pulse Rate
QTc	Corrected QT interval
QTcF	QT interval corrected using Fridericia's formula
QTL	Quality tolerance limit
RR	Respiratory Rate
RSABE	Reference-scaled average bioequivalence

Abbreviation	Definition
SD	Standard Deviation
SADE	Serious adverse device effect
SAE	Serious adverse event
SARS-CoV-2	Severe acute respiratory syndrome-corona virus type 2
SmPC	Summary of product characteristics
SoA	Schedule of activities
Tmax	Time to reach maximum plasma concentration
TEAE	Treatment-emergent adverse event
THC	Tetrahydrocannabinol
ULN	Upper limit of normal
USADE	Unanticipated serious adverse device effect
WHO	World Health Organization
WOCBP	Woman of childbearing potential
WONCBP	Woman of nonchildbearing potential

Term	Definition
Adverse Drug Reaction	An adverse event where a causal relationship between a medicinal product and the adverse event is at least a reasonable possibility, i.e., the relationship cannot be ruled out. a. In the context of a clinical trial, an ADR can be serious or non-serious. Serious ADRs may be subject to expedited reporting if they are considered unexpected (see SUSAR definition). b. For marketed products, ADRs are subject to expedited reporting within the country where they are authorized
Auxiliary Medicinal Product (AxMP) a. Authorized AxMP	Medicinal products used in the context of a clinical trial but not as investigational medicinal products, such as medicinal products used for background treatment, challenge agents, rescue medication, or used to assess endpoints in a clinical trial. Auxiliary medicinal products should not include concomitant medications, that is medications unrelated to the clinical trial and not relevant for the design of the clinical trial. Medicinal product authorized in accordance with Regulation (EC) No 726/2004, or in any Member State concerned in accordance with Directive 2001/83/EC,

Term	Definition
b. Unauthorized AxMP	irrespective of changes to the labeling of the medicinal product. 1. Safety reporting with regard to auxiliary medicinal products shall be made in accordance with Chapter 3 of Title IX of Directive 2001/83/EC. a. Medicinal product not authorized in accordance with Regulation (EC) No 726/2004. 1. Safety reporting for unauthorized auxiliary medicinal products will follow the same processes and procedures as SUSAR safety reporting.
Background treatment	Type of medicinal product administered to each of the clinical trial participant, regardless of randomization group, to treat the indication that is the object of the study. Background treatment is generally considered to be the current standard care for the particular indication. In these trials, the IMP is given in addition to the background treatment and safety efficacy are assessed. The protocol may require that the IMP plus the background treatment is compared with an active comparator or with placebo plus background treatment.
Blinding	A procedure in which 1 or more parties to the study are kept unaware of the intervention assignment in order to reduce the risk of biased study outcomes. The level of blinding is maintained throughout the conduct of the study, 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 SAE. In an open-label study, no blind is used. Both the investigator and the participant know the identity of the intervention assigned.
Certified copy	A copy (irrespective of the type of media used) of the original record that has been verified (i.e., by a dated signature or by generation through a validated process) to have the same information, including data that describe the context, content, and structure, as the original.
Challenge agents	A product given to trial participants to produce a physiological response that is necessary before the pharmacological action of the IMP can be assessed.
Combination product	Combination product comprises any combination of

Term	Definition
	• drug • device • biological product Each drug, device and biological product included in a combination product is a constituent part.
Comparator	Any product used as a reference (including placebo, marketed product, GSK or non-GSK) for an investigational product being tested in a clinical trial. This is any product that is being used to assess the safety, efficacy, or other measurable value against the test product (IMP).
Co-administered (concomitant) products	A product given to clinical trial participants as required in the protocol as part of their standard care for a condition which is not the indication for which the IMP is being tested and is therefore not part of the objective of the study.
Eligible	Qualified for enrollment into the study based upon strict adherence to inclusion/exclusion criteria.
Essential documents	Documents which individually and collectively permit evaluation of the conduct of a study and the quality of the data produced
Intervention Number	A number identifying an intervention to a participant, according to intervention allocation.
Investigational product	A pharmaceutical form of an active substance or placebo being tested or used as a reference in a clinical trial, including products already with a marketing authorization but used or assembled (formulated or packaged) in a way different from the authorized form, or when used for an unauthorized indication, or when used to gain further information about the authorized form.
Investigator	A person responsible for the conduct of the clinical study at a study site. If a study is conducted by a team of individuals at a study site, the investigator is the responsible leader of the team and may be called the principal investigator. The investigator can delegate study-related duties and functions conducted at the study site to qualified

Term	Definition
	individual or party to perform those study-related duties and functions.
Legally acceptable representative	An individual, judicial or other body authorized under applicable law to consent on behalf of a prospective participant to the participant's participation in the clinical study. The terms legal representative or legally authorized representative are used in some settings.
Medicinal products used to assess endpoints	A product given to the participant in a Clinical Trial as a tool to assess a relevant clinical trial endpoint; it is not being tested or used as a reference in the clinical trial.
Participant Number	A unique identification number assigned to each participant who consents to participate in the study.
Participant	Term used throughout the protocol to denote an individual who has been contacted to participate or who participates in the clinical study as a recipient of the study intervention. Synonym: subject
Placebo	An inactive substance or treatment that looks the same as, and is given in the same way as, an active drug or intervention/treatment being studied.
Randomization	Process of random attribution of intervention to participants to reduce selection bias.
Rescue medication	Medicines identified in the protocol as those that may be administered to the participants when the efficacy of the IMP is not satisfactory, or the effect of the IMP is too great and is likely to cause a hazard to the patient, or to manage an emergency situation.
Source data	All information in original records and certified copies of original records of clinical findings, observations, or other activities in a clinical study necessary for the reconstruction and evaluation of the study. Source data are contained in source documents (original records or certified copies).
Standard of Care	Medicine(s) for a specific indication, or a component of the standard care for a particular medical indication,

Term	Definition
	based on national and/or international consensus; there is no regulatory significance to this term. 1. Products/regimens considered standard of care may differ country to country, depending on consensus in individual countries
Study intervention	Term used throughout the clinical study to denote a set of investigational product(s) or marketed product(s) or placebo intended to be administered to a participant. Note: "Study intervention" and "study treatment" are used interchangeably unless otherwise specified.
Study completion date	The date on which the last participant in a clinical study was examined or received an intervention/treatment to collect final data for the primary outcome measures, secondary outcome measures, and AEs (that is, the last participant's last visit or LSLV).
Study monitor	An individual assigned by the sponsor and responsible for assuring proper conduct of clinical studies at 1 or more investigational sites.
SUSAR	Suspected Unexpected Serious Adverse Reaction; in a clinical trial, a serious adverse reaction that is considered unexpected, i.e., the nature or severity of which is not consistent with the reference safety information (e.g., Investigator's Brochure for an unapproved investigational medicinal product). All adverse drug reactions (ADRs) that are both serious and unexpected are subject to expedited reporting

1. PROTOCOL SUMMARY

1.1. Synopsis

Protocol Title:

A Phase 1, randomized, open-label, single dose, 2-treatment arm (200 µg and 800 µg), 4-way cross-over study in healthy participants aged 18 to 55 to compare the pharmacokinetics of salbutamol administered via metered dose inhalers containing propellants HFA-152a (test) and HFA-134a (reference).

Brief Title:

A study to compare the pharmacokinetics of salbutamol administered via metered dose inhalers containing propellants HFA-152a (test) or HFA-134a (reference) in healthy participants aged 18 to 55 inclusive.

Rationale:

In current MDIs, micronized particles are suspended in propellant HFA-134a which has a significant global warming potential. GSK is seeking to develop a low carbon footprint alternative propellant, HFA-152a which will address global climate change. Refer to Section 2.1 for further information.

Objectives, Endpoints, and Estimands:

The primary objective of this study is to characterize the PK of single doses of salbutamol in healthy participants delivered via an MDI containing propellant HFA-152a (test), and to compare with an MDI containing propellant HFA-134a (reference) in Cohort 1 with 200 µg and Cohort 2 with 800 µg. The primary PK endpoints are AUC(0-30min), AUC(0-∞) and Cmax. Refer to Section 3 for further information.

Overall Design:

This a randomized, open-label, single dose, 2-sequence, 2-treatment arm, 4-way cross-over (2x2x4) single-center study in healthy participants aged 18 to 55 years. Salbutamol will be administered as either a 200 µg or 800 µg dose, given as 2 or 8 actuations respectively at 20 second intervals, each delivering 100 µg as the ex-valve dose of HFA-152a or HFA-134a.

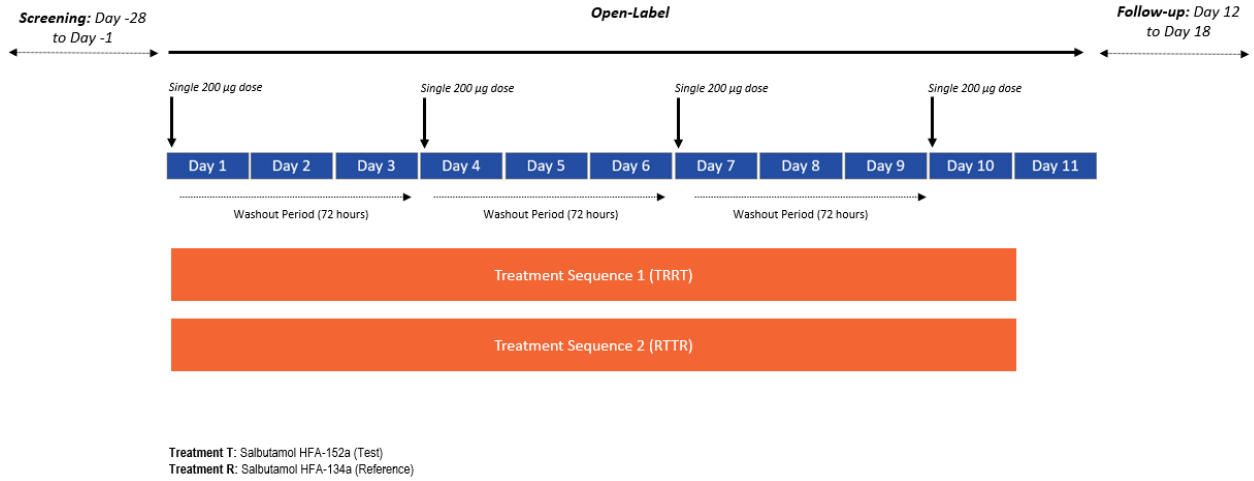
Number of Participants:

The study has planned to enroll a total of 60 participants; 30 participants will be enrolled on to Cohort 1 (200 µg) and 30 participants will be enrolled on to Cohort 2 (800 µg). Within each cohort, 15 participants will be randomized to 1 of the 2 treatment sequences (TRRT and RTTR).

Data Monitoring/Other Committee: Refer to Section 10.1.6.

1.2. Schema

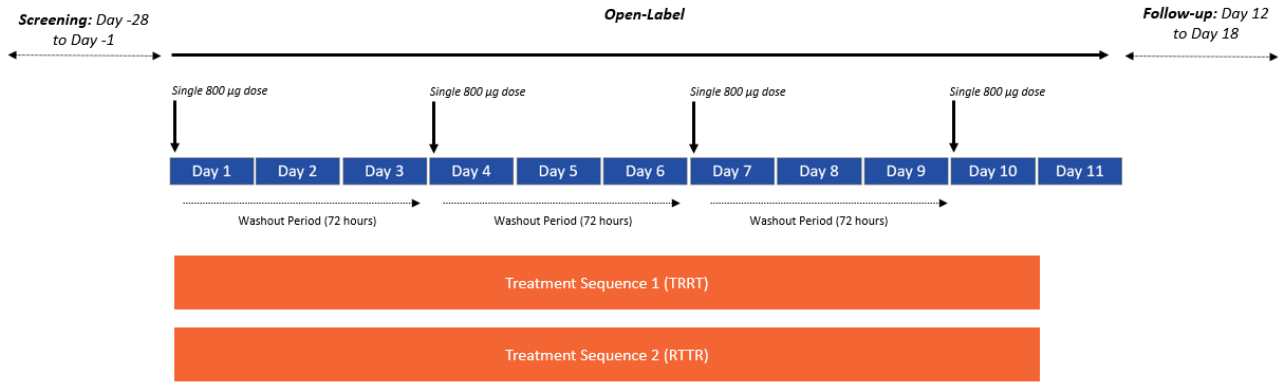
Figure 1 Study Design Overview (Cohort 1 – 200 µg dose)



Treatment T: Salbutamol HFA-152a (Test)
Treatment R: Salbutamol HFA-134a (Reference)

Note. Participants enrolled on to cohort 1 (200 µg) will be randomized to 1 of 2 treatment sequences.

Figure 2 Study Design Overview (Cohort 2 – 800 µg dose)



Treatment T: Salbutamol HFA-152a (Test)
Treatment R: Salbutamol HFA-134a (Reference)

Note. Participants enrolled on to cohort 2 (800 µg) will be randomized to 1 of 2 treatment sequences.

1.3. Schedule of activities (SoA)

Table 1 Schedule of Activities

Procedure	Screening		Intervention period (days)[a]											Discharge or early discontinuation	Follow up[b]
Timepoints	**-28 to -1**	**-1**	**1 (pre-dose)**	**1**	**2**	**3**	**4**	**5**	**6**	**7**	**8**	**9**	**10**	**11**	**12 to 18**
Confinement		X	X	X	X	X	X	X	X	X	X	X	X	X	
Admission		X													
Discharge														X	
Informed consent	X														
Inclusion and exclusion criteria	X	X	X												
Spirometry	X														
Demography	X														
Physical examination[c]	X													X	
Body weight	X														
Height and BMI calculation	X														
Timepoints	**-28 to -1**	**-1**	**1 (pre-dose)**	**1**	**2**	**3**	**4**	**5**	**6**	**7**	**8**	**9**	**10**	**11**	**12 to 18**
Medical history (includes substance usage)	X	X													
Pregnancy test (females only)[d]	X	X												X	

Procedure	Screening		Intervention period (days)[a]											Discharge or early discontinuation	Follow up[b]
FSH and estradiol (females only)	X														
Serology (HBsAg, anti-HCV, anti-HIV 1 and 2)	X														
SARS-CoV-2 PCR test[e]		X													
Cotinine, drug, and alcohol screen	X	X													
Clinical laboratory assessments[f]	X	X												X	
Blood sampling for serum potassium (PD) and glucose (safety)[g]			X	X			X			X			X		
Vital signs[h]	X	X	X	X			X			X			X	X	
Timepoints	**-28 to -1**	**-1**	**1 (pre-dose)**	**1**	**2**	**3**	**4**	**5**	**6**	**7**	**8**	**9**	**10**	**11**	**12 to 18**
12-lead ECG[i]	X	X	X	X			X			X			X	X	
MDI training[j]		X	X				X			X			X		
Randomization			X												
Administration of study intervention[k]				X			X			X			X		
Blood sampling for PK[l]			X	X	X		X	X		X	X		X	X	
AE review[m]				X	X	X	X	X	X	X	X	X	X	X	X
SAE review[m]	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X
Concomitant medication review	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X

a. **Intervention period days:** Participants will be in the clinic for 1 period, from Day-1 until Day 11 (approximately 24 hours after the last dose on Day 10).

b. **Follow-up:** The follow-up period is from Day 12 to Day 18. The minimum follow-up procedures that should be conducted are outlined in the follow-up column. However, further follow-up procedures can be conducted at the discretion of the investigator. **Note.** The final follow-up will be conducted via a telephone call. However, it will be at the discretion of the PI whether a follow-up visit would be required i.e., if further investigation is needed.
c. **Physical examination:** Complete physical examinations will be conducted at screening. Symptom driven physical examinations are conducted at discharge/early withdrawal and may be conducted at any other time, per investigator discretion.
d. **Pregnancy testing:** Female participants will be required to take a serum pregnancy test (Day-1, Screening and Discharge/Early Withdrawal).
e. **SARS-CoV-2 PCR testing:** Sampling of nasal and throat mucosal cells for PCR testing for SARS-CoV-2 may be performed at the discretion of the site based on local guidelines.
f. **Clinical laboratory assessments**: Including clinical chemistry [includes liver chemistries], hematology, and urinalysis will be conducted at screening, on Day-1 (admission), and at discharge.
g. **Blood sampling for serum potassium (PD) and glucose (safety):** On each dosing day (Day 1, 4, 7and 10), blood samples will be taken predose at: 15 and 30 minutes and 1, 1.5, 2 and 4 hours postdose.
h. **Vital signs:** Systolic and diastolic blood pressure, and PR) vital signs will be recorded at screening, admission, on each dosing day (Day 1, 4,7 and 10) at predose and at the following time points postdose: 15, 30 minutes, 1, 2, 4 hours and at discharge. Assessed after a minimum of 5 minutes of the participant resting in the supine position with a completely automated device.
i. **12-lead ECG:** 12-lead ECG will be recorded at screening, admission, on each dosing day (Day 1, 4, 7 and 10) at predose and at: 30 minutes postdose and at discharge. As the ECG parameters QTc and HR will be PD parameters, ECG will be measured in triplicate at predose on Days 1, 4, 7 and 10. The 3 predose measures will be recorded in the eCRF and will be averaged for QTc interval and HR to derive 1 baseline value which will be recorded in the eCRF. Each individual capture of the triplicate 12-lead ECG set should be obtained as closely as possible in succession, but no more than 5 minutes apart from first to third ECG. Postdose 12-lead ECG measurements will be single measurements on all dosing days. Discharge or early discontinuation 12-lead ECG measurements will be single measurements. **Note.** if an ECG meets potential stopping criteria, then the ECG measurement should be repeated in triplicate, with the averaged QT and QTcF used to determine stopping.
j. **MDI training:** Participants will be trained using a placebo HFA-134a propellant via an MDI on Day-1 and prior to dosing on each dosing day (Day 1, 4, 7 and 10). On dosing days, participants should review the instructions use the placebo MDI. The investigator or delegate are to observe the participant performing a single puff using the placebo MDI and provide feedback as necessary until proper technique is observed before conducting the full dose using the investigational inhaler. Additional training using the placebo MDI (e.g., on Day 3) may be performed at the discretion of the site.
k. **Administration of study intervention**: 8 inhalations of 100 µg (800 µg total) of study intervention or 2 inhalations of 100 µg (200 µg total) of study intervention, with the inhalations administered at 20-second intervals.
l. **PK blood sampling:** on each dosing day (Day 1, 4, 7 and 10) PK blood samples will be taken at predose, at 3, 5, 10, 15-, 20-, 30-, and 45-minutes postdose; and at 1, 1.5, 2, 3, 4, 5, 6, 8, 10-, 12-, 16- and 24-hours postdose for plasma salbutamol determination. PK collection will be from the start of study intervention (first inhalation).
m. **AE and SAE review:** AEs will be collected from the start of study intervention until final follow-up telephone call. SAEs will be collected from the signing of the ICF until final follow-up telephone call.

Table 2 Timing of Assessments

Assessment	Timing of Dosing
Study intervention administration	Dosing for each participant will be around the same time (±1 hour) at each intervention period day. All postdose timepoints will be from the start of each study intervention i.e., the first inhalation.
PK blood sampling	Predose samples will be obtained between waking up and dosing. Postdose samples up to 30 minutes will be obtained with a time window of ±1 minute. Thereafter, postdose samples will be obtained with time margins of ±5% of the time that has passed since (last) dosing.
PD blood sampling	For PD blood samples, predose samples will be obtained between waking up and dosing. Postdose samples up to 30 minutes will be obtained with a time window of ±2 minutes. Thereafter, postdose samples will be obtained with time margins of ±5% of the time that has passed since (last) dosing.
Safety assessments	For safety assessments, predose assessments will be performed between waking up and dosing. For safety assessments up to 2.5 hours postdose, a time window of ±15 minutes is allowed. Thereafter, serial postdose assessments (e.g., multiple assessments within any given day) will be performed with time margins of ±10% of the time that has passed since (last) dosing.

Note. When assessments are planned for the same scheme time, the order of the assessments should be arranged in such a way that ECG and vital signs will be done first followed by the PK (then PD) blood sampling, with PK blood sampling exactly on time.

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2. INTRODUCTION

2.1. Study rationale

In the currently marketed Ventolin Evohaler, micronized particles of salbutamol are suspended in propellant HFA-134a [VENTOLIN EVOHALER, 2023]. However, HFA-134a has significant GWP and hence its use in medicine and other applications is being phased out under mounting environmental pressure to replace it with a low carbon footprint alternative such as salbutamol HFA-152a (test).

GSK is seeking to develop a sustainable version of the current Ventolin Evohaler to address global climate change. This study will evaluate the pharmacokinetics of salbutamol administered via MDIs containing propellants HFA-152a (test) and HFA-134a (reference). Hereafter, salbutamol administered via MDIs containing propellant HFA-152a (test) will be referred to as salbutamol HFA-152a and salbutamol administered via MDIs containing propellant HFA-134a (reference) will be referred to as salbutamol HFA-134a.

2.2. Background

Approximately 339 million people currently suffer from asthma, and it is the most common noncommunicable disease among children [GINA, 2023]. Asthma requires medical assessment and care when it occurs on either a persistent, or intermittent but frequently recurring basis. Exacerbations of asthma are episodes characterized by a progressive increase in symptoms of shortness of breath, cough, wheezing, or chest tightness and a progressive decrease in lung function, such that they represent a change from the patient's usual status that is sufficient to require a change in treatment [GINA, 2023] Severe exacerbations are potentially life-threatening and their treatment requires close medical supervision [GINA, 2023].

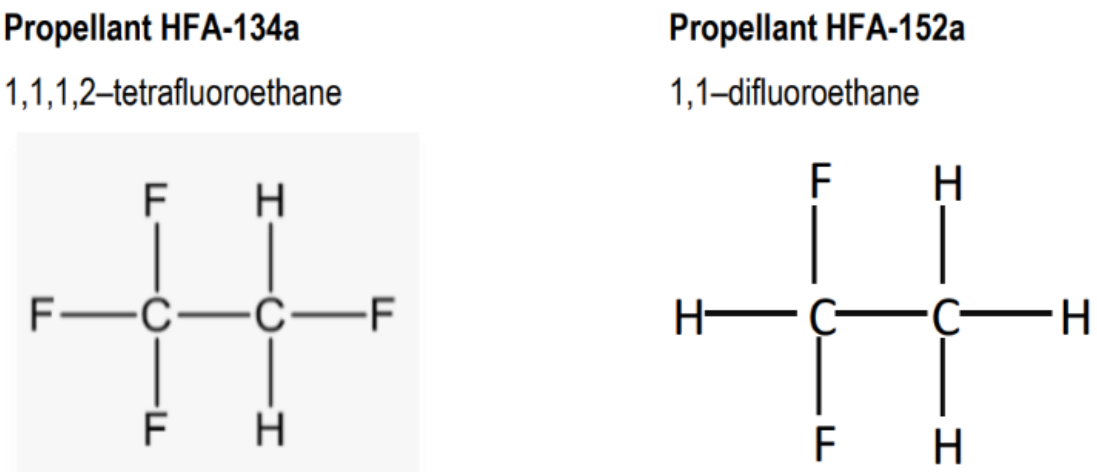
Pharmacotherapy for asthma follows a stepwise process determined by symptoms and disease severity. The most common treatment options are short- and long-acting bronchodilators, corticosteroid anti-inflammatory agents, and combination products containing both bronchodilators and corticosteroids. Biological agents are also available for more severe disease in patients with eosinophilic asthma. Short-acting reliever medications such as salbutamol sulfate are a cornerstone of asthma therapy for as-needed relief of breakthrough symptoms, including worsening asthma or exacerbations [GINA, 2023] Salbutamol sulfate, the active ingredient in Ventolin Evohaler (HFA-134a) is approved in over 130 countries.

Salbutamol sulfate is a selective β2–adrenergic receptor agonist that acts on bronchial smooth muscle providing short-acting (4 to 6 hour) bronchodilation with a fast onset (within 5 minutes) and is suitable for the relief and prevention of asthma symptoms and reversible airway obstruction.

Inhaled salbutamol has been approved for human use since 1968 initially as an MDI containing CFC propellants. These propellants were later replaced with propellant HFA-134a due to international treaties phasing out CFC production arising from environmental

concern. GSK is developing a sustainable version of its Ventolin Evohaler using an alternative new propellant, HFA-152a which will replace the current propellant, HFA-134a. The chemical structures of the 2 propellants are presented in Figure 3.

Figure 3 Chemical structures of HFA-134a and HFA-152a



Salbutamol HFA-152a will be developed as a replacement product for the current Ventolin Evohaler (HFA-134a) with the same indications and posology. HFA-152a is an MDI delivering 100 µg of salbutamol (as salbutamol sulfate) per dose; 200 doses per device. Salbutamol has a duration of action of 4 to 6 hours with administration of up to 2 inhalations (200 µg total) not exceeding 4 times daily.

CCI [redacted]

The maximum recommended total daily dose of salbutamol for chronic use delivered via the Ventolin MDI is 800 µg. This dose requires 8 inhalations (800 µg salbutamol). The exposure to HFA-152a (8x63 µL) is estimated to be 454 µg/day (6.87 µmol/day). This is well below the exposures of HFA-152 shown to have no adverse effects in humans.

2.3. Benefit/risk assessment

Detailed information about the known and expected benefits and risks and reasonably expected AEs of salbutamol delivered via MDI may be found in the patient information, package insert and SmPC. There is no added benefit expected for the healthy participants. A risk assessment has been presented in Section 2.3.1.

2.3.1. Risk assessment

Potential Risk of Clinical Significance	Summary of Data/Rationale for Risk	Mitigation Strategy
Study Intervention		
Participants will be instructed to administer 200 µg or 800 µg of salbutamol. There is a potential risk that the HFA-152a propellant delivers a higher effective dose of salbutamol compared to the reference Ventolin Evohaler	Hypokalemia: β-adrenergic agonist medicines may produce significant hypokalemia in some patients, possibly through intracellular shunting, which has the potential to produce adverse cardiovascular effects. The decrease in serum potassium is usually transient, not requiring supplementation. Hyperglycemia: β-adrenergic agonist medicines may produce a transient rise in serum glucose which is not expected to be clinically significant with inhalational administration. However, large doses of intravenous salbutamol have been reported to aggravate pre-existing diabetes mellitus and ketoacidosis. Cardiovascular effects: β-adrenergic agonist medicines may produce clinically significant cardiovascular effects in some patients such as changes in pulse rate or blood pressure. In addition, β-agonists have been reported to produce ECG changes, such as flattening of the T wave, prolongation of the QTc interval, and ST segment depression. The clinical relevance of these findings is unknown.	Healthy participants are included who will be carefully screened against the defined inclusion and exclusion criteria (including overall cardiac health, vital signs, ECG interval values, and lab safety values [including serum potassium and glucose levels) and by ECG monitoring and lab PD/safety assessments (including serum potassium and glucose levels) as per Section 1.3.
Study Procedures		
There are no risks associated with the study design and/or procedures.		

2.3.2. Benefit Assessment

- The healthy participants will receive medical assessments associated with study procedures, for example, physical examination, ECG and labs. They may, as a result, derive some benefit related to information about their general health status.
- The healthy participants will be contributing to the understanding of PK and PD for all MDI-based therapies with salbutamol and HFA-152a. The healthy participants will be contributing to the process of developing a therapy which has a reduced impact on global warming.

2.3.3. Overall Benefit Risk Conclusion

Taking into account the measures taken to minimize risk to participants participating in this study, the potential risks identified in association with salbutamol administered via MDI containing propellants HFA-152a and HFA-134a are justified by the anticipated benefits to all ongoing and future programs with HFA-152a as a propellant as well as the anticipated environmental and societal benefits of introducing a new propellant (HFA-152a) with greatly reduced GWP compared to the current propellant HFA-134a.

3. OBJECTIVES, ENDPOINTS AND ESTIMANDS

Table 3 Objectives and Endpoints

Objective(s)	Endpoint(s)
Primary	
To characterize the PK of single doses of salbutamol in healthy participants delivered via an MDI containing propellant HFA-152a (test), and to compare with an MDI containing propellant HFA-134a (reference) in the following cohorts: **Cohort 1**: Salbutamol HFA-152a MDI 200 µg (test), HFA-134a MDI 200 µg (reference) **Cohort 2**: Salbutamol HFA-152a MDI 800 µg (test), HFA-134a MDI 800 µg (reference)	• AUC(0-30min) • AUC(0-∞) • Cmax
Secondary	
Pharmacokinetic: To characterize the PK of single doses of salbutamol in healthy participants delivered via an MDI containing propellant HFA-152a (test), and to compare with an MDI containing propellant HFA-134a (reference) in the following cohorts: **Cohort 1:** Salbutamol HFA-152a MDI 200 µg (test), HFA-134a MDI 200 µg (reference) **Cohort 2:** Salbutamol HFA-152a MDI 800 µg (test), HFA-134a MDI 800 µg (reference)	**Pharmacokinetic**: • Tmax • t1/2 • AUC(0-last)

Pharmacokinetics: To measure the intra-participant variability in PK of single doses of salbutamol for healthy participants delivered via an MDI containing propellant in the following cohorts: **Cohort 1**: Salbutamol HFA-152a MDI 200 µg (test), Salbutamol HFA-134a MDI 200 µg (reference) **Cohort 2**: Salbutamol HFA-152a MDI 800 µg (test), Salbutamol HFA-134a MDI 800 µg (reference)	**Pharmacokinetics**: • Intra-participant variability of AUC(0-30min), AUC(0-∞), AUC(0-last), and Cmax of HFA-152a (test) • Intra-participant variability CVw of AUC(0-30min), AUC(0-∞), AUC(0-last), and Cmax of HFA-134a (reference)
Safety: To assess the safety and tolerability of single doses of salbutamol in healthy participants delivered via MDI HFA-152a (test) or propellant HFA-134a (reference) in the following cohorts: **Cohort 1:** Salbutamol HFA-152a MDI 200 µg (test), HFA-134a MDI 200 µg (reference) **Cohort 2**: Salbutamol HFA-152a MDI 800 µg (test), HFA-134a MDI 800 µg (reference)	**Safety**: • Incidence of AEs and SAEs • Absolute values for 12 lead ECGs recording of HR and QTc intervals at each assessed visit • Change from baseline for postdose 12 • lead ECGs recording of HR and QTc intervals at each assessed visit • Absolute values of clinical laboratory parameters at each assessed visit • Absolute values of vital signs (systolic and diastolic blood pressure and pulse rate) at each assessed visit
Exploratory	
Pharmacodynamics: To characterize the PD of single doses of salbutamol in healthy participants delivered via an MDI containing propellant HFA-152a (test), and to compare to an MDI containing propellant HFA-134a (reference) in the following cohorts: **Cohort 1**: Salbutamol HFA-152a MDI 200 µg (test), Salbutamol HFA-134a MDI 200 µg (reference) **Cohort 2**: Salbutamol HFA-152a MDI 800 µg (test), Salbutamol HFA-134a MDI 800 µg (reference)	**Pharmacodynamics**: • Minimum serum potassium • MHR • Maximum QTc interval

3.1. Primary Estimands

Cohort 1:

Test: Salbutamol HFA-152a MDI 200 µg

Reference: Salbutamol HFA-134a MDI 200 µg

The primary clinical question of interest for Cohort 1 is-

What is the GMR for Cohort 1 in primary PK parameters (AUC(0-30min), AUC(0-∞) and Cmax) of salbutamol HFA-152a and salbutamol HFA-134a in healthy participants after administration of single dose of 200 µg?

The primary estimand for Cohort 1 is described by the following attributes:

- The population is healthy male or female participants aged 18 to 55 years.
- The primary PK parameters endpoints are AUC(0-30min), AUC(0-∞) and Cmax.
- Treatment condition is administration of salbutamol as a single 200 µg dose, given as 2 actuations each separated by a 20-second interval, each delivering 100 µg of salbutamol (as salbutamol sulfate) as the ex-valve dose for the HFA-152a MDI or HFA-134a MDI.
- The ICEs and estimand strategies are as follows:
  - Study treatment discontinuation due to any reason (hypothetical strategy).
  - Occurrence of emesis or coughing on dosing (hypothetical strategy).
  - Dosing error: Considered less than or more than prescribed dose (hypothetical
  - strategy).
  - Use of prohibited or rescue medication which impacts to PK parameters (hypothetical strategy).

  For all ICEs, interest lies in evaluating the primary PK parameters and comparisons of the ratio of 2 formulations for each primary PK parameters in a hypothetical scenario in which the participant had not experienced an intercurrent event. This is to minimize potential confounding of ICEs on individual PK parameters across treatment periods.
- The population-level summary to be estimated is:

  Ratio of adjusted GM (for logarithmic transformed values) of 2 formulations with 90% CI for each primary PK endpoint.

Cohort 2:

Test: Salbutamol HFA-152a MDI 800 µg

Reference: Salbutamol HFA-134a MDI 800 µg

The primary clinical question of interest for Cohort 2 is –

What is the GMR for Cohort 2 in primary PK parameters (AUC(0-30min), AUC(0-∞) and Cmax) of salbutamol HFA-152a and salbutamol HFA-134a in healthy participants after administration of single dose of 800 µg?

The estimand for Cohort 2 has the same estimand attributes as Cohort 1 (population, PK endpoints, population-level summary measure, ICE strategy), except for the attribute for the treatment condition:

- Treatment condition is administration of salbutamol as a single 800 µg dose, given as 8 actuations each separated by a 20-second interval, each delivering 800 µg of salbutamol (as salbutamol sulfate) as the ex-valve dose for the HFA-152a MDI or HFA-134a MDI.

Rationale for primary estimands for both cohorts: The test formulation is being developed with an aim to have comparable PK characteristics as the reference formulation. Interest lies in the PK values obtained in the scenarios that had the participant been exposed to correct dose as prescribed, where study treatment discontinuation of the randomized sequence, the occurrence of emesis or coughing on dosing, and prohibited rescue medication usage had not occurred.

3.2. Estimands supporting secondary PK objectives

3.2.1. PK Parameters

Cohort 1:

Test: Salbutamol HFA-152a MDI 200 µg

Reference: Salbutamol HFA-134a MDI 200 µg

The secondary clinical question of interest for Cohort 1 is:

What is the summarized PK profile in Cohort 1 using descriptive statistics for Tmax and t1/2 and GMR for AUC(0-last) between salbutamol HFA-152a and salbutamol HFA-134a in healthy participants after administration of single dose of 200 µg?

The secondary estimand for Cohort 1 has the same estimand attributes as the primary estimand for Cohort 1 (population, treatment condition, ICE strategy), except for the estimand attributes of PK endpoints and population-level summary measure:

- The secondary PK parameters endpoints are Tmax, t1/2, AUC(0-last).
- The population-level summary to be estimated are:
  - AM with 95% CI for Tmax and GM with 95% CI for t1/2.
  - Ratio of adjusted GM (for logarithmic transformed values) of 2 formulations with 90% CI AUC(0-last).

Cohort 2:

Test: Salbutamol HFA-152a MDI 800 µg

Reference: Salbutamol HFA-134a MDI 800 µg

The secondary clinical question of interest for Cohort 2 is –

What is the summarized PK profile in Cohort 2 using descriptive statistics for Tmax and t1/2 and GMR for AUC(0-last) between salbutamol HFA-152a and salbutamol HFA-134a in healthy participants after administration of single dose of 800 µg?

The secondary estimand for Cohort 2 has the same estimand attributes as the primary estimand for Cohort 2 (population, treatment condition, ICE strategy), except for the estimand attributes of PK endpoints and population-level summary measure:

- The secondary PK parameters endpoints are Tmax, t1/2, AUC(0-last).
- The population-level summary to be estimated are:
  - AM with 95% CI for Tmax and GM with 95% CI for t1/2.
  - Ratio of adjusted GM (for logarithmic transformed values) of 2 formulations with 90% CI AUC(0-last).

3.2.2. Intra-participant variability of PK Parameters

Cohort 1:

Test: Salbutamol HFA-152a 200 µg
Reference: Salbutamol HFA-134a 200 µg

The secondary clinical question of interest for Cohort 1 is –

What is the intra-participant variability of the PK profile of Cohort 1 using CVw for AUC(0-30min), AUC(0-∞), Cmax and AUC(0-last) PK parameters for salbutamol HFA-152a and salbutamol HFA-134a in healthy participants after administration of single dose of 200 µg? This secondary objective is to establish whether either the reference or test product is considered a high variable product at the 200 µg dose level (CVw > 30%).

The secondary estimand for Cohort 1 has the same estimand attributes as the primary estimand for Cohort 1 (population, treatment condition, ICE strategy), except for the estimand attributes of PK endpoints and population-level summary measure:

- The secondary PK parameters endpoints for HFA-152a (test) and HFA-134a (reference) are:
  - Intra-participants variability in AUC(0-30min)
  - Intra-participants variability in AUC(0-∞)
  - Intra-participants variability in Cmax
  - Intra-participants variability in AUC(0-last)

- The population-level summary to be estimated for HFA-152a (test) and HFA-134a (reference) are:
  - Intra(within)- participants CVw (%) for AUC(0-30min), AUC(0-∞), Cmax and AUC(0-last)

Cohort 2:

Test: Salbutamol HFA-152a 800 µg

Reference: Salbutamol HFA-134a 800 µg

The secondary clinical question of interest for Cohort 2 is –

What is the intra-participant variability in PK profile of Cohort 2 using CVw for AUC(0-30min), AUC(0-∞), Cmax and AUC(0-last) PK parameters for salbutamol HFA-152a and salbutamol HFA-134a in healthy participants after administration of single dose of 800 µg?. This secondary objective is to establish whether either the reference or test product is considered a high variable product at the 800 µg dose level (CVw > 30%).

The secondary estimand for Cohort 2 has the same estimand attributes as the primary estimand for Cohort 2 (population, treatment condition, ICE strategy), except for the estimand attributes of PK endpoints and population-level summary measure:

- The secondary PK parameters endpoints for HFA-152a (test) and HFA-134a (reference) are:
  - Intra-participants variability in AUC(0-30min)
  - Intra-participants variability in AUC(0-∞)
  - Intra-participants variability in Cmax
  - Intra-participants variability in AUC(0-last)
- The population-level summary to be estimated for HFA-152a (test) and HFA-134a (reference) are:
  - Intra(within)- participants CVw (%) for AUC(0-30min), AUC(0-∞), Cmax and AUC(0-last)

Rationale for supporting secondary estimands of PK parameters for both cohorts: Rational for supporting secondary estimands of PK parameters for both cohorts is same as primary estimands.

3.2.3. Estimands supporting secondary safety objectives

Cohort 1:

Test: Salbutamol HFA-152a MDI 200 µg

Reference: Salbutamol HFA-134a MDI 200 µg

The clinical question of interest for the safety secondary objective in Cohort 1 is –

What is the safety and tolerability profile of single doses 200 µg of salbutamol in Cohort 1 healthy participants delivered via MDIs?

The secondary safety estimand for Cohort 1 is described by the following attributes:

- The population is healthy male or female participants aged 18 to 55 years.
- The secondary safety endpoints are:
  - Incidence of AEs and SAEs
  - Absolute values for predose and postdose 12-lead ECGs recording of HR and QTc intervals at each assessed visit
  - Change from baseline for postdose 12-lead ECGs recording of HR and QTc intervals at each assessed visit
  - Absolute values of clinical laboratory parameters at each assessed visit
  - Absolute values of vital signs (systolic and diastolic blood pressure and pulse rate) at each assessed visit.
- Treatment condition is administration of salbutamol as a single 200 µg dose, given as 2 actuations each separated by a 20-second interval, each delivering 100 µg of salbutamol (as salbutamol sulfate) as the ex-valve dose for the HFA-152a MDI or HFA-134a MDI.
- The ICEs and estimand strategies are as follows:
  - Study treatment discontinuation due to any reason (treatment policy strategy). Interest lies in the group of participants irrespective of whether they completed the study intervention.
  - Occurrence of emesis or coughing on dosing (treatment policy strategy).
  - Dosing error: Considered less than or more than prescribed dose (treatment policy strategy.
  - Use of prohibited or rescue medication which is impacted to PK parameters (Treatment policy strategy).
- The population-level summary to be estimated are:
  - Number and percentages for incidence of AEs and SAEs
  - Mean of absolute values for 12-lead ECGs recording of HR and QTc intervals at each assessed visit
  - Mean of change from baseline values for postdose 12-lead ECGs recording of HR and QTc intervals at each assessed visit
  - Mean of absolute of clinical laboratory parameters including vital signs (systolic and diastolic blood pressure and pulse rate) at each assessed visit

Cohort 2:

Test: Salbutamol HFA-152a MDI 800 µg

Reference: Salbutamol HFA-134a MDI 800 µg

The clinical question of interest for the safety secondary objective in Cohort 2 is –

What is the safety and tolerability profile of single doses 800 µg of salbutamol in Cohort 2 healthy participants delivered via MDIs?

The secondary safety estimand for Cohort 2 has the same estimand attributes as Cohort 1 (population, PK endpoints, population-level summary measure, ICE strategy), except for the attribute for the treatment condition:

- Treatment condition is administration of salbutamol as a single 800 µg dose, given as 8 actuations each separated by a 20-second interval, each delivering 100 µg of salbutamol (as salbutamol sulfate) as the ex-valve dose for the HFA-152a MDI or HFA-134a MDI.

Rationale for estimands supporting secondary safety objectives in both cohorts: This is a study in healthy volunteers with short follow-up duration. Therefore, all adverse events (AEs), serious adverse events (SAEs), 12 lead ECG, laboratory parameters and vital signs data are of interest, regardless of ICEs to support completeness of reporting and transparency of the clinical study.

4. STUDY DESIGN

4.1. Overall design

This is a randomized, open-label, single dose, 2-sequence, 2-treatment arm, 4-way cross-over (2x2x4) single-center study in healthy participants aged 18 to 55 years. Salbutamol will be administered as either a 200 µg or 800 µg dose, given as 2 or 8 actuations respectively at 20 second intervals, each delivering 100 µg as the ex-valve dose of HFA-152a or HFA-134a. Sequential enrollment approach will be followed for the study. Participants in Cohort 2 will be enrolled once participants in Cohort 1 have completed the study. An interim analysis is planned for when last participant enrolled for the 200 µg (Cohort 1) completes their last visit, and final analysis will be conducted when the last participant of 800 µg (Cohort 2) completes their last visit.

The study has planned to enroll a total of 60 participants to either Cohort 1 (200 µg) or Cohort 2 (800 µg) to ensure at least 56 participants in the PK analysis set (see Section 9 on analysis sets). For each cohort, 30 healthy participants will be randomly assigned to a cohort to ensure at least 28 participants are included in the PK analysis set with 15 randomized to each treatment sequence.

Following treatment sequences will be applied in each cohort:

Treatment Sequence 1: TRRT

Treatment Sequence 2: RTTR

Where T is salbutamol HFA-152a (test) and R is salbutamol HFA-134a (reference).

In Cohort 1, salbutamol will be administered as single 200 µg dose, given as 2 actuations each separated by a 20-second interval, each delivering 100 µg of salbutamol (as salbutamol sulfate) as the ex-valve dose for the HFA-152a MDI or HFA-134a MDI.

In Cohort 2, salbutamol will be administered as single 800 µg dose, given as 8 actuations each separated by a 20-second interval, each delivering 100 µg of salbutamol (as salbutamol sulfate) as the ex-valve dose for the HFA-152a MDI or HFA-134a MDI.

Participants will attend a screening visit between Day-28 and Day-1. The intervention period will be 10 days with dosing on Day 1, Day 4, Day 7, and Day 10. Participants will be discharged on Day 11. There will be a follow-up period between Day 12 and Day 18.

4.2. Scientific rationale for study design

Study population

Healthy participants have been chosen as the study population due to the study aims and design and the low risk of clinically significant toxicity at anticipated exposure levels. Additionally, the duration of exposure is sufficiently short to not be able to provide clear therapeutic benefit and justify patients discontinuing current therapies. Moreover, use of healthy participants as opposed to patients will allow a clearer interpretation of the study results, as there will be no confounding factors resulting from changes in disease state and/or concomitant medications.

Cross-Over and Full Replicate Design

A cross-over design to compare the different treatments was chosen to reduce the influence of confounding covariates as each participant serves as their own control. The cross-over design requires a smaller number of participants compared to a parallel group design. In addition, given the broad range of exposures observed for the reference product across historical studies, a full replicate design has been selected whereby the salbutamol HFA-152a and salbutamol HFA-134a are administered twice. The full replicate design chosen can help characterize the extent intra-participant variability impacts the results. If a within-subject variability of >30% is observed for the reference product in this study, comparisons between the test and reference formulations will then be performed using a reference-scaled approach. Carryover effects are expected to be eliminated by introducing a washout phase of a minimum of 72 hours between study drug administrations (expected to be equivalent to at least 5 half-lives of salbutamol).

4.2.1. Participant input into design

Not applicable.

4.3. Justification for dose

Justification for 200 µg dose

The dose of 200 µg was selected as it is the maximal single dose recommended per label and the dose recommended for pharmacokinetic evaluation in the FDA Draft Guidance on Albuterol.

Justification for 800 µg dose

The dose of 800 µg was chosen to ensure adequate plasma concentration-time profile to estimate the AUC(0-∞) CCI [redacted] and minimize within- and between-participant variability in inhaled drug dosing. In addition, the 800 µg dose (8 inhalations) is the maximum recommended daily dose for salbutamol, which has been approved in the EU. CCI [redacted]. Any differences in systemic exposure between the test and reference formulations at this dose level are not expected to increase the risk of known safety concerns with salbutamol, which (as per Section 2.3.1) are being mitigated in any case via careful participant screening and safety assessments.

At the proposed dose of 8 inhalations of salbutamol sulfate, the exposure to HFA-152a (8x63 µL) is estimated to be 454 µg/day (6.87 µmol/day). This is well below the exposures shown to have no adverse effects in humans (see Section 2.2).

Previous study experience

Three previous studies (Study 200921, SALB1002 and SALB1003) used a single dose of 600 µg, 1200 µg and 1200 µg respectively. Study 200921 collected PK samples for 12 hours only, whereas SALB1002 and SALB1003 collected PK samples for 24 hours. CCI [redacted]. Where reported in these studies, treatments were generally well tolerated with no clinically significant laboratory safety abnormalities and no serious adverse events. Adverse events were those expected with high dose β2 agonists but reported in low numbers, these included: tremor, palpitations, tachycardia, flushing and headache which was usually the most frequently reported event.

CCI [redacted]

4.4. End-of-study definition

A participant is considered to have completed the study if the participant has completed all periods of the study including the last visit.

The end of the study is defined as the date of the last visit of the last participant in the study globally.

5. STUDY POPULATION

Prospective approval of protocol deviations to recruitment and enrollment criteria, also known as protocol waivers or exemptions, is not permitted.

5.1. Inclusion criteria

Participants are eligible to be included in the study only if all the following criteria apply:

1. Sex: male or female; females may be of childbearing potential, of nonchild bearing potential, or postmenopausal
2. Age : participants to be 18 to 55 years inclusive, at the time of screening
3. BMI: 18.5 to 29.9 kg/m^2 inclusive, at the time of screening
4. Weight: 45 to 110 kg inclusive, at the time of screening
5. Status: healthy participants
6. At screening and Day-1, females must not be pregnant or lactating, or alternatively must be of nonchildbearing potential.
7. A female participant is eligible to participate if she is not pregnant or breastfeeding, and 1 of the following conditions applies:

- Is a WONCBP as defined in Section 10.4.1.1

OR

- Is a WOCBP and using a contraceptive method that is highly effective, with a failure rate of <1%, as described in Section 10.4.1.1 during the study intervention period and for at least 48 hours after the last dose of study intervention. The investigator should evaluate potential for contraceptive method failure (e.g., noncompliance, recently initiated) in relationship to the first dose of study intervention.
- A WOCBP must have a negative highly sensitive pregnancy test (urine or serum) as required by local regulations) within 7 days before the first dose of study intervention. Please see Section 8.3.5 for further details.
- A serum pregnancy test is required. In such cases, the participant must be excluded from participation if the serum pregnancy result is positive.
- Additional requirements for pregnancy testing during and after study intervention are in Section 8.3.5.
- The investigator/delegate is responsible for review of medical history, menstrual history, and recent sexual activity to decrease the risk for inclusion of a woman with an early undetected pregnancy.

8. All prescribed medication must have been stopped at least 30 days prior to admission to the clinical research center based on investigator judgment. An exception is made for hormonal contraceptives, which may be used throughout the study.
9. All over-the-counter medication, vitamin preparations and other food supplements, or herbal medications (e.g., St. John’s wort) must have been stopped at least 14 days

prior to admission to the clinical research center based on investigator judgment. An exception is made for acetaminophen, which is allowed up to admission to the clinical research center.

10. Ability and willingness to abstain from alcohol from 48 hours (2 days) prior to screening, and from 48 hours (2 days) prior to admission until discharge from the clinical research center.
11. Ability and willingness to abstain from methylxanthine-containing beverages or food (coffee, tea, cola, chocolate, energy drinks) from 48 hours (2 days) prior to admission until discharge from the clinical research center. A serum pregnancy test is required. In such cases, the participant must be excluded from participation if the serum pregnancy result is positive.
12. Good physical and mental health on the basis of medical history, physical examination, clinical laboratory, ECG, and vital signs, as judged by the investigator.
13. Serum potassium and serum glucose levels within reference ranges of the clinical research center.
14. Willing and able to sign the ICF.
15. Spirometry at screening demonstrating FEV1 ≥80% predicted.

5.2. Exclusion criteria

Participants are excluded from the study if any of the following criteria apply:

1. History or presence of cardiovascular, respiratory, hepatic, renal, gastrointestinal, endocrine, hematologic, or neurological disorders capable of significantly altering the absorption, metabolism, or elimination of drugs; constituting a risk when taking the study intervention or interfering with the interpretation of data.
2. History or presence of any form of asthma, including childhood asthma and exercise induced asthma.
3. At screening, systolic blood pressure <90 mmHg or >140 mmHg, or diastolic blood pressure <50 mmHg or >90 mmHg.
4. History of pathological tachycardia, or a pulse rate > 85 bpm at screening or Day-1. **Note.** If the pulse rate is >85 bpm, re-measurement will be allowed up to 2 times during the screening period and Day-1.
5. Lymphoma, leukemia, or any malignancy within the past 5 years except for basal cell or squamous epithelial carcinomas of the skin that have been resected with no evidence of metastatic disease for 3 years.
6. Breast cancer within the past 10 years.
7. A QTcF value of >450 msec at screening based on a triplicate measurement taken at a single timepoint.

NOTES:

- The QTc is the QT interval corrected for heart rate according to Bazett's formula (QTcB), Fridericia's formula (QTcF), and/or another method, machine-read or manually over-read.
- The specific formula that will be used to determine eligibility and discontinuation for an individual participant should be determined prior to initiation of the study and the same formula used throughout the study for an individual participant. In other words, several different formulas cannot be used to calculate the QTc for an individual subject and then the lowest QTc value used to include or discontinue the subject from the trial.

8. Vaccine(s) within 2 weeks prior to admission, or plans to receive such vaccines during the study.
9. Donation or loss of more than 450 mL of blood within 60 days prior to (the first) drug administration. Donation or loss of more than 1.5 L of blood (for male participants) or more than 1.0 L of blood (for female participants) in the 10 months prior to (the first) drug administration in the current study.
10. Participation in a drug study within 30 days prior to (the first) drug administration in the current study. Participation in 4 or more other drug studies in the 12 months prior to (the first) drug administration in the current study.
11. Current enrollment or past participation in this clinical study.
12. ALT >1.5x ULN.
13. Total bilirubin >1.5xULN (isolated total bilirubin >1.5xULN is acceptable if total bilirubin is fractionated and direct bilirubin <35%).
14. Current or chronic history of liver disease or known hepatic or biliary abnormalities (with the exception of Gilbert's syndrome or asymptomatic gallstones).
15. Presence of HBsAg at screening or within 3 months prior to first dose of study intervention.
16. Positive hepatitis C antibody test result at screening or within 3 months prior to first dose of study intervention. NOTE: Participants with positive hepatitis C antibody test result due to prior resolved disease can be enrolled if a confirmatory negative hepatitis C RNA test is obtained.
17. Positive pre-study drug/alcohol screen, including THC.
18. Positive HIV antibody test.
19. Cotinine levels indicative of smoking or history or use of tobacco- or nicotine containing products within 6 months prior to screening. **Note.** Levels of ≥10 ng/mL in urine would exclude a participant.
20. Average intake of more than 24 units of alcohol per week (clinical site standard: 1 unit of alcohol equals approximately 250 mL of beer, 100 mL of wine, or 35 mL of spirits).
21. Regular use of known drugs of abuse, including tetrahydrocannabinol.

22. Use of combustible tobacco products, and non-combustible nicotine delivery systems, inclusive of cigarettes, cigars, pipes, and materials used to "vape" within 6 months prior to screening.
23. Hypersensitivity to any of the study interventions, or components thereof, or drug or other allergy that, in the opinion of the investigator, contraindicates participation in the study.
24. Use of any products intended to treat medical conditions that are not approved by the governing health authority in a given country or region (for example, herbal medicine, health supplements, traditional medicine, homeopathic remedies, etc.).
25. Impairment which would prevent the correct and consistent use of an MDI, as determined by the investigator/delegate.

5.3. Lifestyle considerations

5.3.1. Meals and dietary restrictions

- A fasting period of at least 4 hours is required before obtaining all clinical laboratory blood samples.
- Participants will be advised not to consume any foods containing poppy seeds within 48 hours prior to admission to the clinical research center as this could cause a false positive drug screen result.
- Participants to refrain from Seville oranges, grapefruit or grapefruit juice, pomelos, exotic citrus fruits, grapefruit hybrids, or fruit juices from 7 days before the start of study intervention until after the final dose.
- Meals and snacks (such as decaffeinated coffee, herbal tea, fruit, biscuits) will be provided according to the standard operating procedures of the clinical research center.
- Study intervention will be administered to participants after a light breakfast. Participants will fast for a period of 4 hours after study intervention administration on all dosing days. During fasting, no fluids other than water are allowed; water is allowed ad libitum throughout.

5.3.2. Caffeine, alcohol, and tobacco

- Participants to abstain from ingesting caffeine- or xanthine-containing products (e.g., coffee, tea, cola drinks, chocolate, and energy drinks) for 48 hours prior to admission until after collection of the final PK sample on Day 11.
- Participants to abstain from alcohol for 48 hours prior to admission until after collection of the final PK sample on Day 11.
- Use of tobacco products will not be allowed from 6 months prior to screening until after the final follow-up telephone call.

5.3.3. Activity

- Participants to abstain from strenuous exercise for 96 hours before each blood collection for clinical laboratory tests. Participants may participate in light recreational activities during the study (e.g., watching television, reading)

5.3.4. Other restrictions

- Participants must not donate blood during the study until follow-up telephone call (other than the blood sampling planned for this study).

5.4. Screen failures

A screen failure occurs when a participant who has consented to participate in the clinical study is not subsequently randomized to study intervention. A minimal set of screen failure information is required to ensure transparent reporting of screen failure participants to meet the CONSORT publishing requirements and to respond to queries from regulatory authorities. Minimal information includes demography, screen failure details (screening status, date of screen fail, screen failure rationale), eligibility criteria, and any SAE.

Individuals who do not meet the criteria for participation in this study (screen failure) may be rescreened. Rescreened participants should be assigned a new participant number for every rescreening event. Previously assigned participant numbers are to be recorded in the participants' eCRF.

Note. A participant can only be rescreened once, and the GSK Medical Monitor must be consulted prior to rescreening.

5.5. Criteria for temporarily delaying administration of study intervention

Not applicable.

6. STUDY INTERVENTION(S) AND CONCOMITANT THERAPY

The definition of study intervention is provided in the table of definition.

6.1. Study intervention(s) administered

Table 4 Study Intervention(s) Administered

Intervention Label	Not applicable. Unique identifiers are not used on open-label studies.			
Intervention Name	Salbutamol HFA-152a	Salbutamol HFA-152a	Salbutamol HFA-134a	Salbutamol HFA-134a
Intervention Description	A single 800 µg dose, given as 8 x 100 µg (ex-valve) at 20-second intervals. OR A single 200 µg dose, given as 2 x 100 µg (ex-valve) at 20-second intervals.	A single 800 µg dose, given as 8 x 100 µg (ex-valve) at 20-second intervals. OR A single 200 µg dose, given as 2 x 100 µg (ex-valve) at 20-second intervals.	A single 800 µg dose, given as 8 x 100 µg (ex-valve) at 20-second intervals. OR A single 200 µg dose, given as 2 x 100 µg (ex-valve) at 20-second intervals.	A single 800 µg dose, given as 8 x 100 µg (ex-valve) at 20-second intervals. OR A single 200 µg dose, given as 2 x 100 µg (ex-valve) at 20-second intervals.
Type	Drug	Drug	Drug	Drug
Dose Formulation	Salbutamol sulfate HFA-152a suspension	Salbutamol sulfate HFA-152a suspension	Salbutamol sulfate HFA-134a suspension	Salbutamol sulfate HFA-134a suspension
Unit Dose Strength(s)	100 µg (ex-valve)	100 µg (ex-valve)	100 µg (ex-valve)	100 µg (ex-valve)
Dosage Level(s)	800 µg OR 200 µg	800 µg OR 200 µg	800 µg OR 200 µg	800 µg OR 200 µg
Route of Administration	Inhalation	Inhalation	Inhalation	Inhalation
Use	Investigational Product	Investigational Product	Comparator	Comparator
IMP and NIMP/AxMP.	IMP	IMP	IMP	IMP
Sourcing	Provided centrally by the sponsor	Provided centrally by the sponsor	Provided centrally by the sponsor	Provided centrally by the sponsor

Packaging and Labeling	Study intervention will be provided in a canister fitted with a metering valve within an actuator. Each canister will be labeled as required per country requirement.	Study intervention will be provided in a canister fitted with a metering valve within an actuator. Each canister will be labeled as required per country requirement.	Study intervention will be provided in a canister fitted with a metering valve within an actuator. Each canister will be labeled as required per country requirement.	Study intervention will be provided in a canister fitted with a metering valve within an actuator. Each canister will be labeled as required per country requirement.
Current Name(s) or Alias(es)	--	--	Ventolin	Ventolin

Note. The study has a full replicate design (Test and Reference are replicated twice). Salbutamol HFA-152a and salbutamol HFA-134a are duplicated to reinforce the 4 way cross-over.

Table 5 Study Arm(s)

Arm Title	200 µg	800 µg
Arm Type	Experimental (HFA-152a) and Active Comparator (HFA-134a) administered at 200 µg.	Experimental (HFA-152a) and Active Comparator (HFA-134a) administered at 800 µg.
Arm Description	Participants will receive a single 200 µg dose given as 2x100 µg (ex-valve) at 20-second intervals on Day 1, Day 4, Day 7, and Day 10 of either HFA-152a or HFA-134a.	Participants will receive a single 800 µg dose given as 8x100 µg (ex-valve) at 20-second intervals on Day 1, Day 4, Day 7, and Day 10 of either HFA-152a or HFA-134a.
Associated Intervention Labels	Salbutamol HFA-152a and Salbutamol HFA-134a.	Salbutamol HFA-152a and Salbutamol HFA-134a.

6.1.1. Medical devices

- The GSK manufactured medical devices (or devices manufactured for GSK by a third party) provided for use in this study are MDIs.
- Instructions for medical device use are provided in the pharmacy manual.
- All device deficiencies (including malfunction, use error and inadequate labeling) shall be documented and reported by the investigator throughout the clinical investigation (see Section 8.4.9 and Section 10.6) and appropriately managed by GSK.

Note. MDIs need to be primed before use. The priming of the MDIs should not be done in the vicinity of the participants, as unintended inhalation of micronized particles of salbutamol may affect the salbutamol plasma concentrations of the participants. Please refer to the pharmacy manual for full IFU.

6.2. Preparation, handling, storage, and accountability

- The investigator or designee must confirm appropriate conditions (e.g., temperature) have been maintained during transit for all study intervention received, and any discrepancies are reported and resolved before use of the study intervention.
- Only participants enrolled in the study may receive study intervention, and only authorized site staff may supply, prepare, or administer study intervention.
- For further guidance and information about equipment requirements and product usage, please refer to the pharmacy manual.
- All study intervention must be stored in a secure, environmentally controlled, and monitored (manual or automated) area in accordance with the labeled storage conditions with access limited to the investigator and authorized site staff.
- The investigator, or authorized site staff is responsible for study intervention accountability, reconciliation, and record maintenance (i.e., receipt, reconciliation, and final disposition records)
- Further guidance and information for the final disposition of unused study interventions are provided in the pharmacy manual.
- Under normal conditions of handling and administration, study intervention is not expected to pose significant safety risks to site staff. Take adequate precautions to avoid direct eye or skin contact and the generation of aerosols or mists. In the case of unintentional occupational exposure notify the monitor, medical monitor and/or GSK study contact.
- A MSDS/equivalent document describing occupational hazards and recommended handling precautions either will be provided to the investigator, where this is required by local laws, or is available upon request from GSK.

6.3. Assignment to study intervention

Participants will be independently randomized in Cohort 1 (200 µg) and Cohort 2 (800 µg). Within each cohort, participants will be randomly assigned with 1:1 ratio in 2 possible sequences (TRRT or RTTR [T for test and R for reference]) in 2x2x4 cross-over design as mentioned in Section 4.1.

The randomization schedule, order of treatment sequence assignment, will be generated using the GSK validated randomization system Randall NG. The randomization schedule is made up of randomization numbers mapped to treatment sequences. Randomization numbers are assigned in sequential order. Each participant will be assigned 1 randomization number to determine which treatment sequence will be followed.

Knowledge of treatment assignment will be revealed at time of participant is randomized, but not sooner.

Once the randomization number has been assigned, it must not be re-assigned to another participant.

6.4. Blinding

This is an open-label study; however, the specific intervention to be taken by a participant will be assigned at random, and the order of treatment sequence assignment will remain concealed. Once the randomization is conducted, the treatment sequence assignment will be revealed.

6.5. Study intervention compliance

When participants are dosed at the site, they will receive study intervention directly from the investigator or designee, under medical supervision. The date and time of each dose administered in the clinic will be recorded in the source documents.

In addition, the GSK Medical Monitor may provide training and support to site personnel regarding inhaler technique in order to support study intervention compliance.

Participants will be required to perform predose MDI training as outlined in the SoA. The study will use placebo MDIs so an assessment of co-ordination of inhalation and actuation can be performed, and participants are familiar with the cold freon effect ahead of dosing.

6.6. Dose modification

Not applicable.

6.7. Continued access to study intervention after the end of the study

Not applicable.

6.8. Treatment of overdose

An overdose is any dose of study intervention given to a participant that exceeds the planned, randomized dose for an individual within a given dose group.

GSK does not recommend specific treatment for an overdose.

6.9. Prior and concomitant therapy

Participants must abstain from taking prescription medications from 30 days prior to admission until completion of the follow-up visit or early discontinuation. An exception is made for hormonal contraceptives, which may be used throughout the study.

Participants must abstain from taking nonprescription drugs (including vitamins, recreational drugs, and dietary or herbal supplements) from 14 days prior to admission until completion of the follow-up visit or early discontinuation.

Vaccination is not allowed from 2 weeks prior to admission until discharge.

Paracetamol/Acetaminophen, at doses of ≤2 grams/day, is permitted for use any time during the study, except during the 24 hours predose or postdose. The reason that acetaminophen is not permitted during the 24 hours predose or postdose is because acetaminophen may decrease the excretion rate of salbutamol, which could result in a higher serum level (https://go.drugbank.com/drugs/DB01001). Other concomitant medications may be considered on a case-by-case basis by the investigator in consultation with the medical monitor.

Other medication to treat AEs may only be prescribed if deemed necessary by the investigator. If medication is used, the name of the drug, the dose, and dosage regimen will be recorded in the eCRF.

7. DISCONTINUATION OF STUDY INTERVENTION AND PARTICIPANT DISCONTINUATION/WITHDRAWAL

7.1. Discontinuation of study intervention

‘Discontinuation’ of study intervention refers to any participant who has not received all planned doses of the study intervention. In rare instances, it may be necessary for a participant to permanently discontinue study intervention. If study intervention is permanently discontinued, the participant should, if possible, continue other study procedures (e.g., safety), planned in the study protocol at the discretion of the investigator. If the participant does not agree to continue in-person visits, a modified follow-up must be arranged to ensure the collection of endpoints and safety information (e.g., telephone contact). See the SoA for data to be collected at the time of discontinuation of study intervention and follow-up and for any further evaluations that need to be completed.

The primary reason for premature discontinuation of the study intervention will be documented in the eCRF based on the list below:

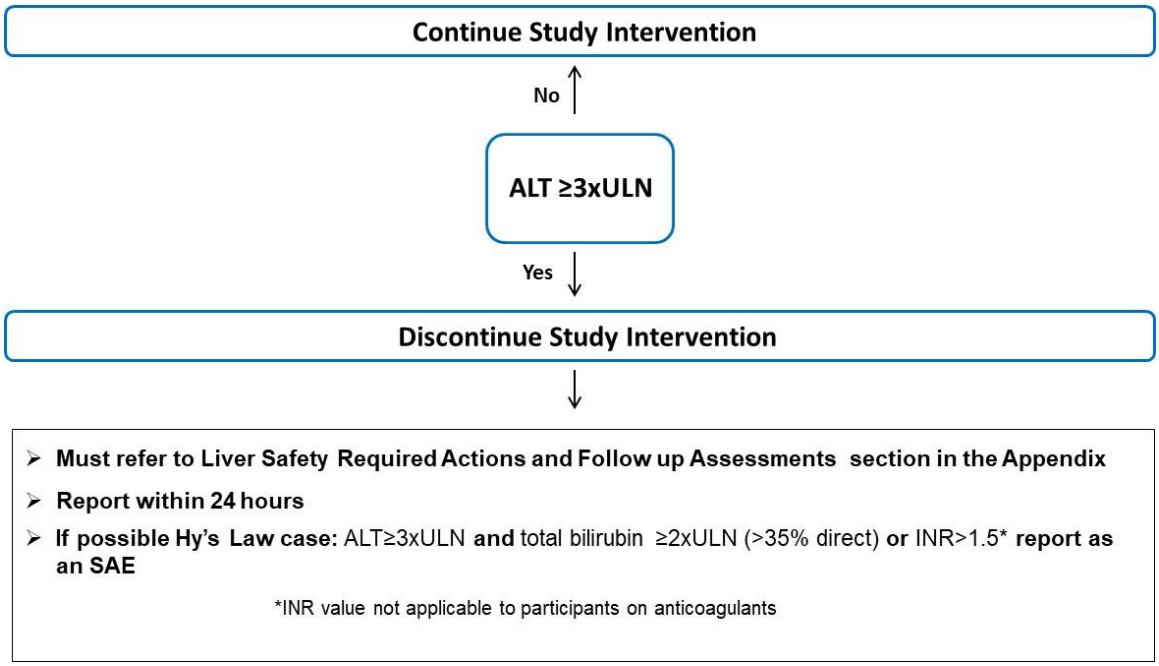
Reasons	Additional items/Sub-reasons
AE	
Lost to follow-up	Participant Relocated Participant was Incarcerated Other Unknown
Participant Reached Protocol-Defined Stopping Criteria	Liver chemistry stopping criteria QTc Stopping criteria
Physician Decision	Specify
Pregnancy	
Protocol Deviation	Specify
Site Terminated by Sponsor	
Sponsor Terminated Study Treatment	
Study Terminated by Sponsor	
Withdrawal by Participant	Burden of Procedure Participant Relocated COVID-19 Pandemic Other
Other	Specify
Death	

7.1.1. Liver chemistry stopping criteria

Discontinuation of study intervention for abnormal liver tests is required by the investigator when a participant meets one of the conditions outlined in Figure 4 or in the presence of abnormal liver chemistries not meeting protocol-specified stopping rules if the investigator believes that it is in best interest of the participant.

Study intervention will be discontinued for a participant if liver chemistry stopping criteria are met:

Figure 4 Phase 1 Liver Chemistry Stopping Criteria – Liver Stopping Event Algorithm



Abbreviations: ALT=alanine transaminase; INR = international normalized ratio; SAE = serious adverse event; ULN = upper limit of normal.

Refer to Section 10.5 for required Liver Safety Actions and Follow-up Assessments.

7.1.2. QTc Stopping criteria

If a clinically significant finding is identified (including, but not limited to, changes from baseline in QT interval corrected using Fridericia's formula [QTcF] after enrollment, the investigator or qualified designee will determine if the participant can continue the study intervention and if any change in participant management is needed. This review of the ECG printed at the time of collection must be documented. Any new clinically relevant finding should be reported as an AE.

A subject that meets the bulleted criteria below based on the average of triplicate ECG readings will be withdrawn from study intervention.

- QTcF > 500 msec
- Change from baseline: QTcF >60 msec
- QT uncorrected > 600 msec

7.2. Participant discontinuation/withdrawal from the study

A participant may withdraw from the study at any time at the participant's own request for any reason (or without providing any reason).

A participant may be withdrawn at any time at the discretion of the investigator for safety, behavioral, or compliance reasons.

Investigators will attempt to contact participants who do not return for scheduled visits or follow-up.

At the time of discontinuing from the study, if possible, an ED visit should be conducted, as shown in the SoA (Section 1.3). See SoA (Section 1.3) for data to be collected at the time of study discontinuation and follow-up and for any further evaluations that need to be completed.

The participant will be permanently discontinued from the study intervention and the study at that time.

All data and samples collected up to and including the date of withdrawal of/last contact with the participant will be included in the study analyses. If the participant withdraws consent for disclosure of future information, the sponsor may retain and continue to use any data collected before such a withdrawal of consent.

If a participant withdraws from the study, the participant may request destruction of any samples taken and not tested, and the investigator must document this in the site study records.

The primary reason for participant discontinuation/withdrawal from the study will be documented in the eCRF based on the list below:

Reasons	Additional items/Sub-reasons
AE	
Lost to follow-up	Participant Relocated Participant was Incarcerated Other Unknown
Participant Reached Protocol-Defined Stopping Criteria	Liver chemistry stopping criteria QTc Stopping criteria
Physician Decision	Specify
Pregnancy	
Protocol Deviation	Specify
Site Terminated by Sponsor	
Study Terminated by Sponsor	
Withdrawal by Participant	Burden of Procedure Participant Relocated COVID-19 Pandemic Other
Other	Specify
Death	

Participants who are withdrawn from the study because of AEs/SAEs must be clearly distinguished from participants who are withdrawn for other reasons. Investigator will follow participants who are withdrawn from the study due to an AE/SAE until the event is resolved (see Section 10.3.4.5)

7.3. Lost to follow-up

A participant will be considered lost to follow-up if the participant repeatedly fails to return for scheduled visits and is unable to be contacted by the study site.

The following actions must be taken if a participant fails to return to the clinic for a required study visit:

1. The site must attempt to contact the participant and reschedule the missed visit as soon as possible, counsel the participant on the importance of maintaining the assigned visit schedule and ascertain whether the participant wishes to and/or should continue in the study.
2. Before a participant is deemed lost to follow-up, the investigator or designee must make every effort to regain contact with the participant (where possible, 3 telephone calls, and if necessary, an email to the participant's last known email address). These contact attempts should be documented in the participant's medical record.
3. Should the participant continue to be unreachable, the participant will be considered to have withdrawn from the study. This scenario will be noted as treatment discontinuation and/or study withdrawal with the primary reason for this being lost to follow-up in the eCRF.

8. STUDY ASSESSMENTS AND PROCEDURES

1. Study procedures and their timing are summarized in the SoA. Protocol waivers or exemptions are not allowed.
2. Adherence to the study design requirements, including those specified in the SoA, is essential and required for study conduct.
3. All screening evaluations must be completed and reviewed to confirm that potential participants meet all eligibility criteria. Subjects who have signed informed consent but are not eligible to proceed should be recorded in the eCRF with a status of 'screen failure'.
4. Procedures conducted as part of the participant's routine clinical management (e.g., blood count) and obtained before signing of the ICF may be utilized for screening or baseline purposes provided the procedures met the protocol-specified criteria and were performed within the timeframe defined in the SoA.
5. In the event of a significant study-continuity issue (e.g., caused by a pandemic), alternate strategies for participant visits, assessments, study intervention distribution and monitoring may be implemented by the sponsor or the investigator, as per local health authority/ethics requirements.

6. The maximum amount of blood collected from each participant over the duration of the study, including any extra assessments that may be required, will not exceed 500 mL.
7. Repeat or unscheduled samples may be taken for safety reasons or for technical issues with the samples.

8.1. Administrative procedures

8.1.1. Collection of demographic data

Record demographic data such as date of birth, sex, race, and ethnicity in the participant's eCRF.

Collection of sex, race and ethnicity data is necessary to assess and monitor the diversity of the trial participants, and to determine if the trial participants are truly representative of the impacted population.

8.1.2. Medical and vaccination history

Obtain the participant's medical/vaccination history by interviewing the participant and/or review of the participant's medical records. Record any pre-existing conditions, signs and/or symptoms present prior to study start in the eCRF.

8.2. Efficacy assessments

Planned timepoints for all efficacy assessments are provided in the SoA (Section 1.3).

MDI Training

MDI training using placebo demonstrator devices will be performed at the timepoints specified in the SoA. The correct inhalation technique will be reinforced by the investigator/delegate prior to treatment administration.

Spirometry

Spirometry should be obtained using spirometry equipment that is calibrated and maintained in accordance with the manufacturers guidance and meets ATS and ERS guidelines [Graham, 2019].

Spirometry will be performed as part of the enrolment criteria where participants pre-bronchodilator FEV1 should be ≥80% of predicted to be eligible to enroll onto the study.

8.3. Safety assessments

Planned timepoints for all safety assessments are provided in the SoA (Section 1.3).

8.3.1. Physical examination

- A complete physical examination will include, at a minimum, assessments of the cardiovascular, respiratory, gastrointestinal, and neurological systems.

8.3.2. Vital signs

- Vital signs that will be measured include pulse rate and blood pressure.
- Blood pressure and pulse measurements will be assessed after a minimum of 5 minutes of the participant resting in the supine position with a completely automated device. Manual techniques will be used only if an automated device is not available.
- If an absolute value for vital signs parameters is clinically significant as judged by the investigator, then the value will be recorded as an AE.

8.3.3. Electrocardiograms

- Twelve lead ECG(s) will be obtained at screening, intervention period, and at discharge or early discontinuation as outlined in the SoA (Section 1.3) using an ECG machine that automatically calculates the heart rate and measures PR, QRS, QT, and QTc intervals.
- The ECG will be obtained after the participant has been resting for at least 5 minutes in the supine position. Refer to Section 7.1.2 for QTc withdrawal criteria and any additional QTc readings that may be necessary.
- If an absolute value for parameters is clinically significant as judged by the investigator, then the value will be recorded as an AE.

8.3.4. Clinical safety laboratory tests

- See Section 10.2 for the list of clinical laboratory tests to be performed in accordance with lab manual and the SoA (Section 1.3).
- The investigator must review the laboratory results, document this review, and record any clinically significant changes occurring during the study as an AE. The laboratory results must be retained with source documents.
- All laboratory tests with values considered clinically significantly abnormal during participation in the study or within 1 day after the last dose of study intervention should be repeated until the values return to normal or baseline or are no longer considered clinically significant by the investigator (who may opt to discuss this with the GSK medical monitor).
  - In the absence of a diagnosis, abnormal laboratory findings assessments or other abnormal results the investigator considers clinically significant will be recorded as an AE or SAE, if they meet the definition of an AE or SAE (refer to Section 10.3.1 and Section 10.3.2).

- If clinically significant values do not return to normal/baseline within a period of time judged reasonable by the investigator, the etiology should be identified, and the sponsor notified.
- If laboratory values from non-protocol-specified laboratory tests performed at the institution's local laboratory require a change in participant management or are considered clinically significant by the investigator e.g., SAE or AE, then the results must be recorded.
- The addresses of clinical laboratories performing the laboratory assessments are documented in the Protocol Supporting Documentation.

8.3.5. Pregnancy testing

- Female participants of childbearing potential must perform a blood pregnancy test before the administration of any dose of study intervention. Pregnancy testing must be done even if the participant is menstruating at the time of the study visit. The study intervention may only be administered if the pregnancy test is negative.
- Refer to Section 8.4.6 for the information on study continuation for participants who become pregnant during the study.
- Refer to Section 5.1 Inclusion Criteria for pregnancy testing entry criteria.
- Pregnancy testing (urine or serum as required by local regulations) should be conducted during study intervention period as detailed in the SoA.
- Pregnancy testing (urine or serum as required by local regulations) should be conducted at the end of relevant systemic exposure.
- Additional serum or urine pregnancy tests may be performed, as determined necessary by the investigator, or required by local regulation, to establish the absence of pregnancy at any time during the participant's participation in the study.

8.4. Adverse Events (AEs) serious adverse events (SAEs), and other safety reporting

For definitions relating to safety information see Section 10.3.

The investigator and any qualified designees are responsible for detecting, documenting, and recording events that meet the definition of an AE or SAE and other safety information and remain responsible for following up all AEs (see Section 8.4.2). This includes events reported by the participant.

The method of recording, evaluating, and assessing causality of AEs and SAEs and the procedures for completing and transmitting SAE reports are provided in Section 10.3.

8.4.1. Time period and frequency for collecting AE, SAE, and other safety information

All SAEs will be collected from the signing of the ICF until final follow-up telephone call at the time points specified in the SoA.

SAEs assessed as related to study participation (e.g., study intervention, protocol-mandated procedures, invasive tests, or change in existing therapy) or related to a GSK product (non-IMP) will be recorded from the time a participant consents to participate in the study.

All AEs will be collected from the start of study intervention until final follow-up telephone call at the timepoints specified in the SoA.

Medical occurrences that begin before the start of study intervention but after obtaining informed consent will be recorded as medical history/current medical conditions, not as AEs.

All SAEs will be recorded and reported to the sponsor or designee immediately and under no circumstance should this exceed 24 hours, as indicated in Section 10.3. The investigator will submit any updated SAE data to the sponsor within 24 hours of it being available.

A poststudy AE/SAE is defined as any event that occurs outside of the AE/SAE reporting period defined in Section 8.4.3.

Investigators are not obligated to actively seek information on AEs or SAEs after conclusion of the study participation. However, if the investigator learns of any SAE, including a death, after a participant has been discharged from the study, the investigator must record it in the medical records. If the investigator considers the event to be reasonably related to the study intervention or study participation, the investigator must promptly notify the sponsor.

8.4.2. Method of detecting AEs and SAEs

Care will be taken not to introduce bias when detecting AEs and SAEs. Open-ended and nonleading verbal questioning of the participant is the preferred method to inquire about AE occurrences.

8.4.3. Follow-up of AEs and SAEs

After the initial AE/SAE report, the investigator is required to proactively follow each participant at subsequent visits/contacts. All SAEs will be followed until resolution, stabilization, the event is otherwise explained, or the participant is lost to follow-up (as defined in Section 7.3). Further information on follow-up procedures is provided in Section 10.3.4.5 and Section 10.6.4.4.

8.4.4. AESIs

Not applicable.

8.4.5. Regulatory reporting requirements for SAEs

- Prompt notification by the investigator to the sponsor of an SAE is essential so that legal obligations and ethical responsibilities toward the safety of participants and the safety of a study intervention under clinical investigation are met. See Section 8.4.3 for reporting timeframes.
- For SAEs, the investigator must always provide an assessment of causality at the time of the initial report, as defined in the Section 10.3.4.3.
- An investigator who receives an investigator safety report describing an SAE or other specific safety information (e.g., summary or listing of SAEs) from the sponsor will review and then file it along with the IB, IFU, or package insert will notify the IRB/IEC, if appropriate according to local requirements.

Table 6 Timeframes for submitting SAE, pregnancy and other events reports to GSK

Type of event	Initial reports		Follow-up of relevant information on a previous report	
	Timeframe	Documents	Timeframe	Documents
SAEs	24 hours* ‡	Paper/Electronic AEs Report	24 hours*	Paper/Electronic AEs Report
Pregnancies	24 hours*	Paper pregnancy notification report	24 hours *	Paper pregnancy follow-up report

* Timeframe allowed after receipt or awareness of the information by the investigator/site staff.
‡ Paper AEs Report will be dated and signed by the investigator (or designee). For each SAE, the investigator(s) must document in the medical notes that they have reviewed the SAE and have provided an assessment of causality.

8.4.6. Pregnancy

Female participants who become pregnant after the first study intervention dose must not receive subsequent doses of the study intervention but may continue other study procedures at the discretion of the investigator.

- Details of all pregnancies in female participants will be collected after the start of study intervention and until 48 hours after the last study intervention.
- If a pregnancy is reported, the investigator will record pregnancy information on the appropriate form and submit it to the sponsor within 24 hours of learning of the female participant pregnancy.
- Any pregnancy or pregnancy complication or elective termination of a pregnancy for medical reasons will be reported as an AE or SAE.
- Abnormal pregnancy outcomes (e.g., spontaneous abortion, fetal death, stillbirth, congenital anomalies, ectopic pregnancy) are considered SAEs and will be reported as such.
- The participant will be followed to determine the outcome of the pregnancy. The investigator will collect follow-up information on the participant/pregnant female partner and the neonate, and the information will be forwarded to the sponsor. See Table 6 for reporting timeframes.

- Any poststudy pregnancy-related SAE considered reasonably related to the study intervention by the investigator will be reported to the sponsor as described in Section 8.4.4. While the investigator is not obligated to actively seek this information in former study participants, he or she may learn of an SAE through spontaneous reporting.
- Any female participant who becomes pregnant while participating in the study will discontinue study intervention or be withdrawn from the study.

8.4.7. CV and Death Events

CV Events

Not applicable.

Death Events

The Death eCRF is provided immediately after the occurrence or outcome of death is reported. Initial and follow-up reports regarding death must be completed within 1 week of when the death is reported.

8.4.8. Disease-related events and/or disease-related outcomes not qualifying as AEs or SAEs

Not applicable.

8.4.9. Contact information for reporting SAEs and pregnancies

Table 7 Contact information for reporting SAEs and pregnancies

Study contact for questions regarding SAEs and pregnancies Contact GSK's local and/or medical contacts
Contacts for reporting SAEs and pregnancies. Available 24/24 hours and 7/7 days fax # +44(0) 20 81814780. Available during working hours oax37649@gsk.com

8.4.10. Participant card

Not applicable.

8.4.11. Medical device deficiencies

Medical devices are being provided for use in this study as the study intervention. To fulfill regulatory reporting obligations worldwide, the investigator is responsible for the

detection and documentation of events meeting the definitions of device deficiency that occur during the study with such devices.

The definition of a medical device deficiency can be found in Section 10.6.

NOTE: Deficiencies fulfilling the definition of an AE/SAE will follow the processes outlined in Section 10.6 of the protocol.

8.4.11.1. Time Period for Detecting Medical Device Deficiencies

- Medical device deficiencies that result in an incident will be detected, documented, and reported during all periods of the study in which the medical device is used.
- If the investigator learns of any device deficiency at any time after a participant has been discharged from the study, and such a deficiency is considered reasonably related to a medical device provided for the study, the investigator will promptly notify the sponsor.

The method of documenting medical device deficiencies is provided in Section 10.6.

8.4.11.2. Follow-up of Medical Device Deficiencies

- Follow-up applies to all participants, including those who discontinue study intervention.
- The investigator is responsible for ensuring that follow-up includes any supplemental investigations as indicated to elucidate the nature and/or causality of the deficiency.
- New or updated information will be recorded on the originally completed form with all changes signed and dated by the investigator.

8.4.11.3. Prompt Reporting of Device Deficiencies to the Sponsor

- Device deficiencies will be reported to the sponsor within 24 hours after the investigator determines that the event meets the protocol definition of a medical device deficiency.
- The medical device deficiency report form will be sent to the sponsor by email/facsimile equipment. If email/facsimile equipment is unavailable, then notification by telephone with a copy of data collection tool sent by overnight courier service should be utilized.
- The sponsor will be the contact for the receipt of device deficiency reports.

8.4.11.4. Regulatory Reporting Requirements for Device Deficiencies

- The investigator will promptly report all device deficiencies occurring with any medical device provided for use in the study in order for the sponsor to fulfill the legal responsibility to notify appropriate regulatory authorities and other entities about certain safety information relating to medical devices being used in clinical studies.

- The investigator, or responsible person according to local requirements (e.g., the head of the medical institution), will comply with the applicable local regulatory requirements relating to the reporting of device deficiencies to the IRB/IEC.

8.5. Pharmacokinetics

Whole blood samples of approximately 4.5 mL will be collected for measurement of plasma concentrations of salbutamol as specified in the SoA (Section 1.3).

CCI [redacted]

Instructions for the collection and handling of plasma samples will be provided by the sponsor. The actual date and time (24-hour clock time) of each sample will be recorded.

Details of PK blood sampling collection, processing, storage, and shipping procedures are provided in the Lab Manual prepared by GSK.

8.6. Pharmacodynamics

The following PD parameters will be evaluated and compared: serum potassium weighted means and minimum serum potassium.

Blood samples of approximately 3.5 mL will be collected for measurement of potassium concentrations in serum as specified in the SoA (Section 1.3).

Serum potassium concentrations will be assessed by the CCI [redacted]

8.7. Genetics

Not applicable.

8.8. Biomarkers

Not applicable.

8.9. Immunogenicity assessments

Not applicable.

8.10. Health economics or medical resource utilization and health economics

Neither health economics or medical resource utilization and health economics parameters are evaluated in this study.

9. STATISTICAL CONSIDERATIONS

The statistical analysis plan will include a more technical and detailed description of the statistical analyses described in this section. This section is a summary of the planned statistical analyses of the most important endpoints including primary and key secondary endpoints.

Any deviation from the statistical analysis plan will be reported in the section “Changes in Planned Analysis” in the CSR.

9.1. Statistical hypotheses

This study is designed to estimate the relative bioavailability of salbutamol HFA-152a (test) and Salbutamol HFA-134a (reference) in following cohorts:

Cohort 1: Salbutamol HFA-152a 200 µg, Salbutamol HFA-134a 200 µg.

Cohort 2: Salbutamol HFA-152a 800 µg, Salbutamol HFA-134a 800 µg.

In each cohort, for each PK parameters endpoints (AUC(0-30min.), AUC(0-∞), Cmax and AUC(0-last)), point estimates and corresponding 90% CIs will be constructed for GMR(T/R).

90% Confidence Interval of GMR (T/R): The GMR is calculated by taking the ratio of the geometric means of the salbutamol HFA-152a (T) to the salbutamol HFA-134a (R). The 90% confidence interval of this GMR is a range that provides a measure of the uncertainty in this ratio.

An additional assessment will be performed by comparing the 90% CIs for the GMR(T/R) in AUC(0-30min), AUC(0-∞), Cmax and AUC(0-last) PK parameters to the bioequivalence acceptance range (refer to Section 10.8, Table 12).

9.1.1. Multiplicity Adjustment

No multiplicity adjustment will be performed for the primary or secondary endpoints.

9.2. Analysis sets

Analysis Set	Definition/Criteria	Analyses Evaluated
Screened	• All participants who were screened for eligibility.	All Screened, Reason for Screen Failures
Enrolled	• All participants who screen passed and entered in the study (who were randomized). • Note screening failures (who never passed screening even if rescreened) and participants screened but never enrolled into the study (met eligibility but not needed) are excluded from the • enrolled analysis set as they did not enter the study.	Study Population
FAS	• All randomized participants who received at least a single dose period of study intervention • Data will be reported according to the randomized study intervention.	Exploratory PD objective
Safety	• All participants who received at least one puff/actuation of study intervention.	Estimand for secondary safety objective
PK	• All participants in the FAS who had at least 1 non-missing PK assessment (NQ values will be considered as non-missing values). • Data will be reported according to the actual study intervention.	Estimand for primary and secondary objective of PK parameters

9.3. Statistical analyses

9.3.1. General considerations of pharmacokinetic data

PK analysis will be the responsibility of CCI [redacted] Plasma concentration-time data will be analyzed by noncompartmental methods with WinNolin 8.3. Calculations will be based on the actual sampling times recorded during the study.

The PK parameters data will be summarized using descriptive statistics (N, n, AM, 95% CI of AM, GM, 95% CI of GM, SD, SD (loge) median, between- subject coefficient of variation (CVb in %), minimum and maximum), and will be listed and summarized in tabular and/or graphical form.

Listings and graphical presentation will also be produced for PK parameters endpoints.

Discrete data will be summarized by counts and percentages as appropriate.

9.3.1.1. Definition of PK endpoints

The definitions of PK parameters are described in Table 8.

Table 8 Pharmacokinetic Parameters

Parameters	Descriptions
AUC(0-30min)	Area under the plasma concentration-time curve up to 30 minutes postdose
AUC(0-∞)	Area under the plasma concentration-time curve from time 0 to infinity calculated as: AUC(0-∞) = AUC(0-last)+C(last)/k(el), where C_{last} is the last measurable plasma concentration and K(el) is the elimination rate constant
AUC(0-last)	Area under the plasma concentration-time curve up to last time with concentrations above the LLOQ
Cmax	Maximum observed plasma concentration
Tmax	Time to Cmax
t1/2	Apparent terminal phase half-life

9.3.1.2. Main Analytical Approach

The primary analysis for PK parameters AUC(0-30min), AUC(0-∞) and Cmax will evaluate the primary estimand in PK analysis set.

If for the reference product, SD_{WR} <0.294 or CVw <30%, then ABE approached will be used, refer Section 10.8, Table 12 for the acceptance range limit of BE.

If for the reference product, $SD_{WR} \geq$ 0.294 or CVw ≥30%, then RSABE approached will be used, refer Section 10.8, Table 12 for the acceptance range limit of BE.

The extent of the widening is defined based upon the within-subject variability seen in the bioequivalence study using scaled-average-bioequivalence according to [U, L]=exp [$\pm k \cdot S_{WR}$], where U is the upper limit of the acceptance range, L is the lower limit of the acceptance range, k is the regulatory constant set to 0.760 as per EMA and 0.893 according to FDA and S_{WR} is the within-subject standard deviation of the log-transformed values of Cmax and AUC of the reference product (refer Section 10.8, Table 12).

9.3.1.3. Statistical model for primary analysis

For each cohort, following $\log_e$-transformation, AUC(0-30min.), AUC(0-∞) and Cmax will be analyzed using mixed effect model approach with fixed effect for period, treatment group and participants as random effects. The point estimates and their associated 90% CI will then be back-transformed to provide point estimates and 90% CI for the ratios (GMR) of HFA-152a MDI (test)/HFA-134a MDI (reference).

If an intercurrent event (ICE) (as described in Section 3.1) happens during a specific treatment period, and this event affects the measurement of individual pharmacokinetic (PK) concentrations (pharmacokinetic data), then the data points for those affected individuals in the treatment period will be set to missing.

9.3.1.4. Secondary endpoint(s) analyses

The secondary analysis for PK parameters AUC(0-30min), AUC(0-∞) and Cmax will evaluate the secondary estimand in PK analysis set.

In each cohort, analytical approach for the statistical analysis of AUC(0-last) is same as primary PK parameters as mentioned in Section 9.3.1.2.

Statistical model for estimating the intra-participants (within) variability.

For each cohort, following $\log_e$-transformation, AUC(0-30min.), AUC(0-∞), Cmax and AUC(0-last) will be analyzed using mixed effect model approach with fixed for period, treatment group and participants as random effects. SD in log scale and coefficient of variation (CV_{WR})for intra (within)-participants will be computed.

Statistical model for estimating the inter-participants (intra participants for test and reference) variability.

For each cohort, following $\log_e$-transformation, AUC(0-30min.), AUC(0-∞), Cmax and AUC(0-last) will be analyzed using mixed effect model approach with fixed for period, treatment group and treatment group as random effects. SD in log scale and coefficient of variance (CV) for inter-participants will be calculated.

- Total participants variability will be calculated as sum of intra (within)-participants and inter-participants variability.

If an intercurrent events (ICEs) (as described in Section 3.2.1 and Section 3.2.2) happens during a specific treatment period, and this event affects the measurement of individual pharmacokinetic (PK) concentrations (pharmacokinetic data), then the data points for those affected individuals in the treatment period will be set to missing.

9.3.1.5. Safety analyses

All safety analyses will be performed on the Safety analysis set.

Safety and tolerability will be assessed through AEs, SAEs, clinical laboratory, vital signs, and ECGs, and any other parameter that is relevant for safety assessment. Safety data will be presented in tabular and/or graphical format and summarized descriptively according to GSK's IDSL standards.

Treatment policy strategy for the ICEs (as described in Section 3.2.3) will be used for summarizing safety data of the study.

Safety analysis is described in further detail in below sections.

9.3.1.6. Adverse Events

All reported AEs will be coded using the standard GSK dictionary, (MedDRA) and grouped by body system and preferred terms. Counts and percentages for AEs, and SAEs will be produced by system organ class and preferred terms. AEs and SAEs will also be presented for both the overall follow-up period and on-treatment period. The number and percentages will be provided for all AEs, study treatment – related AEs, and SAEs, deaths, fatal AEs, non-fatal SAEs, and AEs leading to study treatment or study withdrawal. AEs and SAEs will be summarized by severity.

9.3.1.7. Clinical laboratory

A summary of all data outside the reference range of the clinical laboratory will be provided. Absolute values of clinical laboratory data will be presented descriptively (arithmetic mean, SD, median, minimum, and maximum), where applicable.

9.3.1.8. Vital signs and electrocardiograms

Absolute values for vital signs (systolic and diastolic blood pressure and pulse rate) will be summarized descriptively (arithmetic mean, SD, median, minimum, and maximum).

Absolute and changes from baseline values for ECG parameters detailed in Section 8.3.3 will be presented descriptively (arithmetic mean, SD, median, minimum, and maximum), where applicable.

9.3.2. Tertiary/exploratory endpoint(s) analysis

9.3.2.1. Pharmacodynamic analysis

For each cohort, the following PD parameters will be evaluated and compared between Salbutamol HFA-152a MDI (test) and Salbutamol HFA-134a MDI (reference): minimum serum potassium, MHR and maximum QTcF interval.

Following $\log_e$-transformation, minimum serum potassium, MHR and maximum QTcF interval will be analyzed using mixed effect model approach with fixed effect for period, treatment group and participants as random effects. The point estimates and their associated 90% CI will then be back-transformed to provide point estimates and 90% CI for the ratios (GMR) of HFA-152a MDI (test)/HFA-134a MDI (reference).

The exploratory PD parameters data will be summarized using descriptive statistics (N, n, AM, GM, SD, SD in log scale CV %, median, minimum, and maximum), and will be summarized in tabular and/or graphical form.

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[redacted]

9.5. Sample size determination

Up to 60 healthy participants will be enrolled to either Cohort 1 (200 µg) or Cohort 2 (800 µg) to achieve at least 56 participants in the PK analysis set.

For each cohort, 30 healthy participants will be randomly assigned to a cohort to ensure at least 28 participants are included in the PK analysis set with 15 randomized to each treatment sequence.

The target number of participants in each cohort is chosen to achieve half width (refer Table 9) of the 90% CI, in the estimate of the geometric mean ratios for the primary endpoints (AUC (0-30 min.), AUC(0-inf) and Cmax).

In the previously conducted 219430 pilot PK study, there was considerable interest in AUC(0-30 min) due to larger than expected observed differences between test and reference product, in addition to observing potentially higher amounts of intra-subject variability that would classify the product as highly variable (~30%). Though, less variability was observed among AUC(0-inf) and Cmax. Due to uncertainty of what the true CVw may be for both the reference and test products, this current study therefore has considered a more conservative assumption accounting for up to 50% intra subject variability (CVw) in PK parameters, with a particular interest in AUC(0-30 min).

Therefore, a sample size 28 in each cohort, assuming intra subject variability (CVw) of 50% in AUC(0-30 min.), the half width of the upper bound of the 90% CI for observed GMR (T/R) is 16.44% (refer Table 9).

For example, if the geometric ratio between test and reference is 1.06 for n=28 and CVw = 50%, then the upper bound of the 90% CI for observed treatment ratio is 1.06*(1+0.1644) = 1.23 and the lower bound is 1.06/(1+0.1644) = 0.91 (refer Table 10).

Table 9 Sample sizes with varying intra subject variability and associated half-widths of the 90% CI of the observed treatment ratio

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[redacted]

CCI

[redacted]

Table 10 Sensitivity Table

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[redacted]

10. SUPPORTING DOCUMENTATION AND OPERATIONAL CONSIDERATIONS

10.1. Appendix 1: Regulatory, ethical, and study oversight considerations

10.1.1. Regulatory and ethical considerations

- This study will be conducted in accordance with the protocol and with the following:
  - Consensus ethical principles derived from international guidelines including the Declaration of Helsinki and CIOMS international ethical guidelines.
  - Applicable ICH GCP guidelines
  - Applicable laws and regulations

- The protocol, protocol amendments, ICF, IB, and other relevant documents (e.g., advertisements) must be submitted to an IRB/IEC by the investigator and reviewed and approved by the IRB/IEC before the study is initiated.
- Substantial amendments to the protocol will require IRB/IEC approval before implementation of changes made to the study design, except for changes necessary to eliminate an immediate hazard to study participants.
- Protocols and any substantial amendments to the protocol will require health authority approval prior to initiation except for changes necessary to eliminate an immediate hazard to study participants.
- The investigator will be responsible for the following, as applicable:
  - Providing written summaries of the status of the study to the IRB/IEC annually or more frequently in accordance with the requirements, policies, and procedures established by the IRB/IEC
  - Notifying the IRB/IEC of SAEs or other significant safety findings as required by IRB/IEC procedures.
- Providing oversight of the conduct of the study at the site and adherence to requirements of 21 CFR, ICH guidelines, the IRB/IEC, European regulation 536/2014 for clinical studies (if applicable), European Medical Device Regulation 2017/745 for clinical device research (if applicable), and all other applicable local regulations.

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[redacted]

10.1.3. Informed consent process

- The investigator or the investigator's representative will explain the nature of the study, including the risks and benefits, to the participants and answer all questions regarding the study.
- Potential participants must be informed that their participation is voluntary. They will be required to physically or digitally sign a statement of informed consent that meets the requirements of 21 CFR 50, local regulations, ICH guidelines, HIPAA requirements, privacy and data protection requirements, where applicable, and the IRB/IEC or study center.
- The medical record must include a statement that physical or digital informed consent was obtained before the participant was enrolled in the study and the date the physical or digital consent was obtained. The authorized person obtaining the informed consent must also sign the ICF.
- Participants must be reconsented to the most current version of the ICF(s) during their participation in the study.

- A physical or digital copy of the ICF(s) must be provided to the participant.

In case of unexpected pregnancy, participant must be informed that PI such as date of birth, sex of the baby will be collected as part of safety follow-up. Consent for the baby may be obtained from the participant and/or their partner as per local regulations.

If partners of male participants become pregnant during the study, consent will need to be obtained or notification given as per local regulation to the partner before collecting their PI such as LMP, year of birth or the PI such as date of birth, sex of their baby as part of safety follow-up.

10.1.4. Recruitment strategy

Participants will be identified for potential recruitment using clinical database, and IEC/IRB-approved newspaper/radio/social media advertisements, prior to consenting to take part in this study.

10.1.5. Data protection

- Participants will be assigned a unique identifier by the investigator. Any participant records or datasets that are transferred to the sponsor will contain the identifier only; participant names or any information which would make the participant identifiable will not be transferred.
- GSK will ensure protection of the personal data of the investigator and site staff which is collected within the framework of and for the purpose of the study.
- The participant must be informed that their personal study-related data will be used by the sponsor in accordance with local data protection law. The level of disclosure must also be explained to the participant that their data will be used as described in the informed consent.
- The participant must be informed that their medical records may be examined by Clinical Quality Assurance auditors or other authorized personnel appointed by the sponsor, by appropriate IRB/IEC members, and by inspectors from regulatory authorities.
- The contract between sponsor and study sites specifies responsibilities of the parties related data protection, including handling of data security breaches and respective communication and cooperation of the parties.

Information technology systems used to collect, process, and store study-related data are secured by technical and organizational security measures designed to protect such data against accidental or unlawful loss, alteration, or unauthorized disclosure or access.

10.1.6. Committees structure

Not applicable.

10.1.7. Dissemination of Clinical Study Data

- The key design elements of this protocol and results summaries will be posted on www.ClinicalTrials.gov and/or GSK Clinical Study Register in compliance with applicable regulations/GSK policy. GSK will aim to register protocols summaries prior to study start and target results summaries submission within 12 months of primary/study completion date. Where external regulations require earlier disclosure, GSK will follow those timelines.
- Where required by regulation, summaries will also be posted on applicable national or regional clinical study registers.
- Where required by applicable regulatory requirements, an investigator signatory will be identified for the approval of the study report, and provided reasonable access to statistical tables, figures, and relevant reports. GSK will also provide the investigator with the full summary of the study results, including a summary of trial results understandable to laypersons. The investigator is encouraged to share the plain language summary with the study participants, as appropriate. The full study report will be made available upon request, after decision on marketing authorization by regulatory authorities.
- GSK will provide the investigator with the randomization codes and participant-level line listings for their site only after completion of the full statistical analysis.
- GSK intends to make anonymized participant-level data from this study available to external researchers for scientific analyses or to conduct further research that can help advance medical science or improve patient care. This helps ensure the data provided by study participants are used to maximum effect in the creation of knowledge and understanding.

10.1.8. Data quality assurance

- All participant data relating to the study will be recorded on printed or electronic CRFs unless transmitted to the sponsor or designee electronically (e.g., laboratory data). The investigator is responsible for verifying that data entries are accurate and correct by physically or electronically signing the CRF.
- Guidance on completion of eCRFs will be provided in eCRF completion guidelines.
- The investigator must permit study-related monitoring, audits, IRB/IEC review, and regulatory agency inspections and provide direct access to source documents.
- QTLs will be predefined in the Action Item Key Decision Log to identify systematic issues that can impact participant right, safety and/or reliability of study results. These predefined parameters will be monitored during the study, and important deviations from the QTLs and remedial actions taken will be summarized in the CSR.
- Monitoring details describing strategy, including definition of study critical data items and processes (e.g., risk-based initiatives in operations and quality such as risk management and mitigation strategies and analytical risk-based monitoring, involvement of central reading mechanism) methods, responsibilities, and

requirements, including handling of noncompliance issues and monitoring techniques (central, remote, or on-site monitoring) are provided in the monitoring plan.

- The sponsor or designee is responsible for the data management of this study, including quality checking of the data.
- The sponsor assumes accountability for actions delegated to other individuals (e.g., contract research organizations).
- Records and documents, including signed ICF, pertaining to the conduct of this study must be retained by the investigator for 25 years from the issue of the final CSR/equivalent summary unless local regulations or institutional policies require a different retention period. No records may be destroyed during the retention period without the written approval of the sponsor. No records may be transferred to another location or party without written notification to the sponsor.
- When copies of source documents are shared externally for review by a central reader mechanism (e.g., endpoint adjudication committee; expert reader), documents are stored by the external body for 25years.

10.1.9. Source documents

- Source documents provide evidence for the existence of the participant and substantiate the integrity of the data collected. Source documents are filed at the investigator's site.
- Data reported on the eCRF or entered in the eCRF that are transcribed from source documents must be consistent with the source documents or the discrepancies must be explained. The investigator may need to request previous medical records or transfer records, depending on the study. Also, current medical records must be available.
- Definition of what constitutes source data and its origin can be found in source data acknowledgment or monitoring guidelines.
- The investigator must maintain accurate documentation (source data) that supports the information entered in the eCRF.
- The sponsor or designee will perform monitoring to confirm that data entered into the eCRF by authorized site personnel are accurate, complete, and verifiable from source documents; that the safety and rights of participants are being protected; and that the study is being conducted in accordance with the currently approved protocol and any other study agreements, ICH GCP, and all applicable regulatory requirements.

10.1.10. Study and site start and closure

Start of study and first act of recruitment

The start of study and the first act of recruitment are defined as FSFV (first ICF signature date).

Study/Site Termination

GSK or designee reserves the right to close the study site or terminate the study at any time for any reason at the sole discretion of GSK. Study sites will be closed upon study

completion. A study site is considered closed when all required documents and study supplies have been collected and a study site closure visit has been performed.

The investigator may initiate study site closure at any time, provided there is reasonable cause and sufficient notice is given in advance of the intended termination.

Reasons for the early closure of a study site by the sponsor or investigator may include but are not limited to:

For study termination:

- Discontinuation of further study intervention development

For site termination:

- Failure of the investigator to comply with the protocol, the requirements of the IRB/IEC or local health authorities, the sponsor's procedures, or GCP guidelines
- Inadequate or no recruitment (evaluated after a reasonable amount of time) of participants by the investigator
- Total number of participants included earlier than expected

If the study is prematurely terminated or temporarily suspended, the sponsor shall promptly inform the investigators, the IECs/IRBs, the regulatory authorities, and any contract research organization(s) used in the study of the reason for termination or temporary suspension, as specified by the applicable regulatory requirements. The investigator shall promptly inform the participant and should assure appropriate participant therapy and/or follow-up.

10.1.11. Publication policy

The results of this study may be published in peer reviewed scientific literature and/or presented at scientific meetings. The sponsor will comply with the requirements for publication of study results in accordance with standard editorial and ethical practice and as per the sponsor's internal policy. Authorship will be determined by mutual agreement and in line with ICMJE authorship requirements.

10.2. Appendix 2: Clinical laboratory tests

- The tests detailed in Table 11 will be performed by CCI [redacted]
- Local laboratory results are only required in the event that CCI [redacted] results are not available in time for either study intervention administration and/or response evaluation. If a local sample is required, it is important that the sample for central analysis is obtained at the same time. Additionally, if the local laboratory results are used to make either a study intervention decision or response evaluation, the results must be recorded.
- Protocol-specific requirements for inclusion or exclusion of participants are detailed in Section 5 of the protocol.
- Additional tests may be performed at any time during the study as determined necessary by the investigator or required by local regulations.
- Investigators must document their review of each laboratory safety report.

Table 11 Protocol-required safety laboratory tests

Laboratory Tests	Parameters	
Hematology	• Platelet count	
	• Red blood cell (RBC) count	
	RBC indices	• Mean corpuscular volume (MCV) • Mean corpuscular hemoglobin (MCH) • %Reticulocytes
	Absolute WBC count with differential:	• Neutrophils • Lymphocytes • Monocytes • Eosinophils • Basophils
	• **Hemoglobin**	
	• **Hematocrit**	
Clinical chemistry[1]	• Blood urea nitrogen (BUN)/Urea • Potassium** • Creatinine* • Sodium • Calcium • Glucose fasting** • CPK	• Aspartate aminotransferase (AST)/serum glutamic-oxaloacetic transaminase (SGOT) • Alanine aminotransferase (ALT)/serum glutamic-pyruvic transaminase (SGPT) • Alkaline phosphatase • Total bilirubin • Direct bilirubin • Total protein

Laboratory Tests	Parameters
Routine urinalysis	• Specific gravity • pH, glucose, protein, blood, ketones, [bilirubin, urobilinogen, nitrite, leukocyte esterase] by dipstick • Microscopic examination. Please note, this will only be performed if there is an abnormality in accordance with Clinical Laboratory standard procedures. • Microscopic examination (if blood or protein is abnormal)
Pregnancy testing	• Highly sensitive serum hCG pregnancy test.
Other screening tests	• FSH and estradiol. • Cotinine, alcohol, and drug screen (to include at minimum: amphetamines [including XTC], methadone, barbiturates, cocaine, opiates, cannabinoids and benzodiazepines) • Serology [(HIV antibody 1/2 antibody test, hepatitis B surface antigen [HBsAg], and hepatitis C virus antibody)] • PCR testing for SARS-CoV-2 may be performed at the discretion of the physician and based on local guidelines.

NOTES

1. Details of liver chemistry stopping criteria and required actions and follow-up are given in Section [7.1.1 Liver Chemistry Stopping Criteria] and Section10.5: Liver Safety: Suggested Actions and Follow-up Assessments [and Study Intervention Rechallenge Guidelines]]. All events of ALT [or AST] ≥3 × upper limit of normal (ULN) and total bilirubin ≥2 × ULN (> 35% direct bilirubin) or ALT [or AST] ≥3 × ULN and international normalized ratio (INR) > 1.5 (if INR measured), which may indicate severe liver injury (possible Hy's law), must be reported to [sponsor] in 24 hours (excluding studies of hepatic impairment or cirrhosis).

* To assess the kidney function, use eGFR 2021 calculator (CKD-Epi creatinine equation). eGFR (based on CKD-Epi) will be measured at all time points when creatinine is measured.

** Blood sampling for serum potassium and glucose on each dosing day (Day 1, 4, 7 and 10) and at the following time points: predose and 15 and 30 minutes, 1, 1.5, 2 and 4 hours post dose.

10.3. Appendix 3: AEs and SAEs: Definitions and procedures for recording, evaluating, follow-up, and reporting

10.3.1. Definition of AE

AE definition
• An AE is any untoward medical occurrence in a clinical study participant, temporally associated with the use of a study intervention, whether or not considered related to the study intervention. • NOTE: An AE can therefore be any unfavorable and unintended sign (including an abnormal laboratory finding), symptom, or disease (new or exacerbated) temporally associated with the use of a study intervention.
Events Meeting the AE Definition
• Any abnormal laboratory test results (hematology, clinical chemistry, or urinalysis) or other safety assessments (e.g., ECG, radiological scans, vital signs measurements), including those that worsen from baseline, considered clinically significant in the medical and scientific judgment of the investigator (i.e., not related to progression of underlying disease). • Exacerbation of a chronic or intermittent pre-existing condition including either an increase in frequency and/or intensity of the condition. • New condition detected or diagnosed after study intervention administration even though it may have been present before the start of the study. • Signs, symptoms, or the clinical sequelae of a suspected intervention- intervention interaction. • Signs, symptoms, or the clinical sequelae of a suspected overdose of either study intervention or a concomitant medication. Overdose per se will not be reported as an AE/SAE unless it is an intentional overdose taken with possible suicidal/self-harming intent. Such overdoses should be reported regardless of sequelae. • Events that occur as a result of protocol-mandated procedures (i.e., invasive procedures, modification of participant's previous therapeutic regimen).
Events <u>NOT</u> Meeting the AE Definition
• Any clinically significant abnormal laboratory findings or other abnormal safety assessments that are associated with the underlying disease, unless judged by the investigator to be more severe than expected for the participant's condition. • Medical or surgical procedure (e.g., endoscopy, appendectomy): the condition that leads to the procedure is the AE. • Situations in which an untoward medical occurrence did not occur (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 diseases will be recorded in the medical history section of the eCRF.
- Hospitalization for elective treatment of a pre-existing condition (known or diagnosed before signing the informed consent) that did not worsen from baseline.

10.3.2. Definition of SAE

An SAE is defined as any untoward medical occurrence that, at any dose, meets one or more of the criteria listed:

a. Results in death

b. Is life-threatening

The term 'life-threatening' in the definition of 'serious' refers to an event in which the participant was at risk of death at the time of the event. It does not refer to an event, which hypothetically might have caused death, if it were more severe.

c. Requires inpatient hospitalization or prolongation of existing hospitalization

- In general, hospitalization signifies that the participant has been admitted (usually involving at least an overnight stay) at the hospital or emergency ward for observation and/or treatment that would not have been appropriate in the physician’s office or outpatient setting. Complications that occur during hospitalization are AE. If a complication prolongs hospitalization or fulfills any other serious criteria, the event is serious. When in doubt as to whether “hospitalization” occurred or was necessary, the AE should be considered serious.
- Hospitalization for elective treatment of a pre-existing condition that did not worsen from baseline is not considered an AE.

d. Results in persistent or significant disability/incapacity

- The term disability means a substantial disruption of a person’s ability to conduct normal life functions.
- This definition is not intended to include experiences of relatively minor medical significance such as uncomplicated headache, nausea, vomiting, diarrhea, influenza, and accidental trauma (e.g., sprained ankle) that may interfere with 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 participant

f. Abnormal pregnancy outcomes (e.g., spontaneous abortion, fetal death, stillbirth, congenital anomalies, ectopic pregnancy)

g. Is a suspected transmission of any infectious agent via an authorized medicinal product.

h. Other situations:

- Possible Hy's Law case: ALT ≥3x ULN AND total bilirubin ≥ 2x ULN (>35% direct bilirubin) or INR >1.5 must be reported as SAE
- Medical or scientific judgment should be exercised by the investigator in deciding whether SAE reporting is appropriate in other situations such as significant medical events that may jeopardize the participant or may require medical or surgical intervention to prevent one of the other outcomes listed in the above definition. These events should usually be considered serious.
  - Examples of such events include invasive or malignant cancers, intensive treatment for allergic bronchospasm, blood dyscrasias, convulsions, or development of intervention dependency or intervention abuse.

10.3.3. Definition of TEAE

TEAE Definition:

- A TEAE is an event that emerges during treatment, having been absent pre-treatment or worsens relative to the pre-treatment state.

10.3.4. Recording, assessment and follow-up of AE, SAE, and pregnancies

10.3.4.1. AE and SAE recording

- When an AE/SAE occurs, it is the responsibility of the investigator to review all documentation (e.g., hospital progress notes, laboratory reports, and diagnostics reports) related to the event.
- The investigator will then record all relevant AE/SAE information.
- It is not acceptable for the investigator to send photocopies of the participant's medical records to GSK in lieu of completion of the required form.
- There may be instances when copies of medical records for certain cases are requested by GSK. In this case, all participant identifiers, with the exception of the participant number, will be redacted on the copies of the medical records before submission to GSK.
- The investigator will attempt to establish a diagnosis of the event based on signs, symptoms, and/or other clinical information. Whenever possible, the diagnosis (not the individual signs/symptoms) will be documented as the AE/SAE.

10.3.4.2. Assessment of intensity

The investigator will make an assessment of intensity for each AE, SAE and device deficiency reported during the study and assign it to one of the following categories:

- Mild:
  A type of AE that is usually transient and may require only minimal treatment or

therapeutic intervention. The event does not generally interfere with usual activities of daily living.

- Moderate:
  A type of AE that is usually alleviated with additional specific therapeutic intervention. The event interferes with usual activities of daily living, causing discomfort but poses no significant or permanent risk of harm to the research participant.
- Severe:
  A type of AE that interrupts usual activities of daily living, or significantly affects clinical status, or may require intensive therapeutic intervention.

10.3.4.3. Assessment of causality

- The investigator is obligated to assess the relationship between study intervention and each occurrence of each AE/SAE/device deficiency. The investigator will use clinical judgment to determine the relationship.
- A reasonable possibility of a relationship conveys that there are facts, evidence, and/or arguments to suggest a causal relationship, rather than a relationship cannot be ruled out.
- Alternative causes, such as underlying disease(s), concomitant therapy, and other risk factors, as well as the temporal relationship of the event to study intervention administration, will be considered and investigated.
- For causality assessment, the investigator will also consult the IB and/or product information, for marketed products.
- The investigator must review and provide an assessment of causality for each AE/SAE and document this in the medical notes There may be situations in which an SAE has occurred and the investigator has minimal information to include in the initial report to GSK. However, it is very important that the investigator always makes an assessment of causality for every event before the initial transmission of the SAE data to GSK.
- The investigator may change their opinion of causality in light of follow-up information and send an SAE follow-up report with the updated causality assessment.
- The causality assessment is one of the criteria used when determining regulatory reporting requirements.

10.3.4.4. Assessment of outcomes

The investigator will assess the outcome of all serious and non-serious unsolicited AEs recorded during the study as:

- Recovered/resolved
- Recovering/resolving
- Not recovered/not resolved

- Recovered with sequelae/resolved with sequelae
- Fatal (SAEs only).

10.3.4.5. Follow-up of AEs, SAEs, pregnancies or any other events of interest

- The investigator is obligated to perform or arrange for the conduct of supplemental measurements and/or evaluations as medically indicated or as requested by GSK to elucidate the nature and/or causality of the AE or SAE as fully as possible. This may include additional laboratory tests or investigations, histopathological examinations, or consultation with other health care professionals.
- If a participant dies during participation in the study or during a recognized follow-up period, the investigator will provide GSK with a copy of any postmortem findings including histopathology.
- New or updated information will be recorded in the originally submitted documents.
- The investigator will submit any updated SAE data to GSK within 24 hours of receipt of the information.

After the initial AE/SAE/pregnancy or any other event of interest, the investigator is required to proactively follow each participant at subsequent visits/contacts. All SAEs, will be followed until the event is resolved, stabilized, otherwise explained, or the participant is lost to follow-up.

Other non-serious AEs must be followed until the participant is lost to follow-up.

Follow-up during the study

AEs documented at a previous visit/contact and defined as not recovered/not resolved or recovering/resolving will be reviewed at subsequent visits/contacts until follow-up visit or until the participant is lost to follow-up.

If a participant dies during their participation in the study or during a recognized follow-up period, GSK will be provided with any available postmortem findings, including histopathology.

Follow-up of pregnancies

Pregnant participants 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 using the paper pregnancy follow-up report and the AE Report if applicable. Generally, the follow-up period does not need to be longer than 6 to 8 weeks after the estimated date of delivery.

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

Furthermore, the investigator must report any SAE occurring as a result of a poststudy pregnancy that is considered by the investigator to be reasonably related to the study intervention, to GSK as described in the Section 10.3.4.7.

10.3.4.6. Updating of SAE and pregnancy information after removal of write access to the participant's eCRF

When additional SAE or pregnancy information is received after write access to the participant's eCRF is removed, 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 sent to the Study contact for reporting SAEs (refer to Section 8.4.1).

10.3.4.7. Reporting of SAEs and pregnancies

SAE Reporting to GSK via an Electronic Data Collection Tool

- The primary mechanism for reporting an SAE to GSK will be the electronic data collection tool.
- If the electronic system is unavailable, then the site will use the paper SAE data collection tool (see next section) to report the event within 24 hours.
- The site will enter the SAE data into the electronic system as soon as it becomes available.
- After the study is completed at a given site, the electronic data collection tool will be taken offline to prevent the entry of new data or changes to existing data.
- If a site receives a report of a new SAE from a study participant or receives updated data on a previously reported SAE after the electronic data collection tool has been taken offline, then the site can report this information on a paper SAE form (see next section) or to the medical monitor by telephone.
- If the site during the course of the study or poststudy becomes aware of any serious, non-serious AEs, pregnancy exposure, related to any GSK non-IMP they will report these events to GSK or to the concerned CA via the national spontaneous reporting system. These will be classified as spontaneous ICSRs.
- Contacts for SAE reporting can be found in Section 8.4.9.

SAE Reporting to GSK via Paper Data Collection Tool

- Email/facsimile transmission of the SAE paper data collection tool is the preferred method to transmit this information to the medical monitor.
- In rare circumstances and in the absence of email/facsimile equipment, notification by telephone is acceptable with a copy of the SAE data collection tool sent by overnight mail or courier service.
- Initial notification via telephone does not replace the need for the investigator to complete and sign the SAE data collection tool within the designated reporting timeframes.
- Contacts for SAE reporting can be found in Section 8.4.9.

10.4. Appendix 4: Contraceptive and barrier guidance

10.4.1. Definitions

10.4.1.1. Woman of Childbearing Potential (WOCBP)

Women in the following categories are considered WOCBP (fertile):

1. Adolescents of childbearing potential: Tanner stage ≥2 (post-thelarche) irrespective of the occurrence of menarche or following menarche.
2. From the time of menarche until becoming postmenopausal unless permanently sterile (see below)

Note: Menarche is the first onset of menses in a young female. Menarche is normally preceded by several changes associated with puberty including breast development and pubic hair growth.

10.4.1.2. Woman of Nonchildbearing Potential (WONCBP)

Women in the following categories are considered WONCBP:

1. Premenarchal: Tanner stage 1 (prepubertal)
2. Permanently sterile due to one of the following procedures:

a. Documented hysterectomy

b. Documented bilateral salpingectomy

c. Documented bilateral oophorectomy

For permanently sterile individuals due to an alternate medical cause other than the above, (e.g., Mullerian agenesis, androgen insensitivity, gonadal dysgenesis), investigator discretion should be applied to determining study entry. If reproductive status is questionable, additional evaluation should be considered.

Note: Documentation can come from the site personnel's review of the participant's medical records, medical examination, or medical history interview.

3. Postmenopausal female

A postmenopausal state is defined as no menses for 12 months without an alternative medical cause.

- A high FSH level in the postmenopausal range may be used to confirm a postmenopausal state in women not using hormonal contraception HRT. However, in the absence of 12 months of amenorrhea, confirmation with more than one FSH measurement threshold if required (>40 IU/L or mIU/mL) or remove to allow for flexibility with different local thresholds for defining postmenopausal state is required.

- Females on HRT and whose menopausal status is in doubt must discontinue HRT to allow confirmation of postmenopausal status before study enrollment.

10.4.2. Contraception guidance

Female participants of childbearing potential with fertile male partners must use one of the following contraceptive methods from at least 4 weeks prior to first study intervention until 48 hours postdose of last study intervention.

- Abstinence, defined as sexual inactivity consistent with the preferred and usual lifestyle of the participant. Periodic abstinence (e.g., calendar, ovulation, symptothermal, post-ovulation methods) and withdrawal are not acceptable methods of contraception.
- Use of a highly effective contraceptive (see list below) plus partner use of a condom.

• **CONTRACEPTIVES[a] ALLOWED DURING THE STUDY INCLUDE:**
• **Highly Effective Methods[b] That Have Low User Dependency** Failure rate of <1% per year when used consistently and correctly.
Implantable progestogen-only hormone contraception associated with inhibition of ovulation[c]
Intrauterine device (IUD)
Intrauterine hormone-releasing system (IUS)[c]
Bilateral tubal occlusion/ligation
Azoospermic partner (vasectomized or due to a medical cause) • Azoospermia is a highly effective contraceptive method provided that the partner is the sole sexual partner of the woman of childbearing potential and the absence of sperm has been confirmed. If not, an additional highly effective method of contraception should be used. Spermatogenesis cycle is approximately 90 days. Note: documentation of azoospermia for a male participant can come from the site personnel's review of the participant's medical records, medical examination, or medical history interview.
• **Highly Effective Methods[b] That Are User Dependent** Failure rate of <1% per year when used consistently and correctly.
Combined (estrogen- and progestogen-containing) hormonal contraception associated with inhibition of ovulation[c] • oral • intravaginal • transdermal • injectable

Progestogen-only hormone contraception associated with inhibition of ovulation[c] • oral • injectable
Sexual abstinence • Sexual abstinence is considered a highly effective method only if defined as refraining from heterosexual intercourse during the entire period of risk associated with the study intervention. The reliability of sexual abstinence needs to be evaluated in relation to the duration of the study and the preferred and usual lifestyle of the participant.
a. Contraceptive use by men or women should be consistent with local regulations regarding the use of contraceptive methods for those participating in clinical studies. a. Failure rate of <1% per year when used consistently and correctly. Typical use failure rates differ from those when used consistently and correctly. b. Male condoms must be used in addition to hormonal contraception. If locally required, in accordance with Clinical Trial Facilitation Group (CTFG) guidelines, acceptable contraceptive methods are limited to those which inhibit ovulation as the primary mode of action. Note: Periodic abstinence (calendar, sympto-thermal, post-ovulation methods), withdrawal (coitus interruptus), spermicides only, and lactational amenorrhea method (LAM) are not acceptable methods of contraception. Male condom and female condom should not be used together (due to risk of failure from friction)

10.5. Appendix 5: Liver safety: suggested actions and follow-up assessments

Phase 1 Liver chemistry stopping criteria have been designed to assure subject safety and to evaluate liver event etiology.

Phase 1 Liver Chemistry Stopping Criteria and Required Follow-Up Assessments

<table>
<tr><th colspan="2">Liver Chemistry Stopping Criteria</th></tr>
<tr><td>ALT-absolute</td><td>ALT≥3xULN
If ALT≥3xULN AND total bilirubin≥2xULN (>35% direct bilirubin) or international normalized ratio (INR) >1.5, report as an SAE[1,2].</td></tr>
<tr><th colspan="2">Required Actions, Monitoring and Follow-up Assessments</th></tr>
<tr><th>Actions</th><th>Follow-Up Assessments</th></tr>
<tr><td>• Immediately discontinue study intervention
• Report the event to GSK within 24 hours
• Complete the liver event form and complete an SAE data collection tool if the event also meets the criteria for an SAE[2]
• Perform liver event follow-up assessments as described in the Follow-Up Assessment column
• Do not restart or rechallenge participant with study intervention
• Monitor the participant until liver chemistries resolve, stabilize, or return to within baseline (see MONITORING)

MONITORING:
If ALT≥3xULN AND total bilirubin ≥ 2xULN or INR >1.5:</td><td>• Viral hepatitis serology[3]
• Obtain INR and recheck with each liver chemistry assessment until the aminotransferases values show downward trend
• Obtain blood sample for pharmacokinetic (PK) analysis, within 72 hours of identified liver event[4]
• Obtain serum CPK and LDH, GGT, glutamate dehydrogenase (GLDH) and serum albumin.
• Fractionate bilirubin if total bilirubin ≥2xULN
• Obtain complete blood count with differential to assess eosinophilia
• Record the appearance or worsening of clinical symptoms of liver injury, or hypersensitivity, on the liver event form
• Record use of concomitant medications on the concomitant medications report form including acetaminophen, herbal remedies, recreational drugs, and other over-the-counter medications
• Record alcohol use on the liver event alcohol intake form

If ALT≥3xULN AND total bilirubin ≥2xULN</td></tr>
</table>

Liver Chemistry Stopping Criteria	
• Repeat liver chemistries (include ALT, aspartate transaminase [AST], alkaline phosphatase, total bilirubin and INR) and perform liver event follow-up assessments within 24 hours • Monitor participant twice weekly until liver chemistries resolve, stabilize or return to within baseline • A specialist or hepatology consultation is recommended **If ALT≥3xULN AND total bilirubin < 2xULN and INR ≤1.5:** • Perform liver chemistries (include ALT, AST, alkaline phosphatase, total bilirubin and INR) and perform liver event follow-up assessments within 24-72 hours • Monitor participant weekly until liver chemistries resolve, stabilize or return to within baseline	**or INR >1.5 obtain the following in addition to the assessments listed above:** • Anti-nuclear antibody, anti-smooth muscle antibody, Type 1 anti-liver kidney microsomal antibodies, and quantitative total immunoglobulin G (IgG) or gamma globulins • Liver imaging (ultrasound, magnetic resonance, or computed tomography) and /or liver biopsy to evaluate liver disease, complete Liver Imaging form. • Liver biopsy may be considered and discussed with local specialists if available, for instance: – In participants when serology raises the possibility of autoimmune hepatitis (AIH) – In participants when suspected DILI progresses or fails to resolve on withdrawal of study intervention – In participants with acute or chronic atypical presentation. • If liver biopsy is conducted, then complete liver biopsy form

1. Serum bilirubin fractionation should be performed if testing is available. If serum bilirubin fractionation testing is unavailable, record the absence/presence of detectable urinary bilirubin on dipstick, which is indicative of direct bilirubin elevations suggesting liver injury.
2. All events of ALT 3xULN and total bilirubin 2xULN (>35% direct bilirubin) or ALT 3xULN and INR >1.5, which may indicate severe liver injury (possible 'Hy's Law'), must be reported as an SAE, the INR threshold value stated will not apply to participants receiving anticoagulants.
3. Includes: hepatitis A immunoglobulin (IgM) antibody; HBsAg and HBcAb; hepatitis C RNA; cytomegalovirus IgM antibody; Epstein-Barr viral capsid antigen IgM antibody (or if unavailable, heterophile antibody or monospot testing) and hepatitis E IgM antibody
4. Record the date/time of the PK blood sample draw and the date/time of the last dose of study intervention prior to PK blood sample draw. If the date or time of the last dose is unclear, provide the participant's best approximation. If the date/time of the last dose cannot be approximated OR a PK sample cannot be collected in the time period indicated above, do not obtain a PK sample. Instructions for sample handling and shipping are in the lab manual.

10.6. Appendix 6: Medical device AEs, ADEs, SAEs, sADEs, USADEs and device deficiencies: Definitions and procedures for recording, evaluating, follow-up, and reporting in medical device studies

- Both the investigator and the sponsor will comply with all local reporting requirements for medical devices.
- The detection and documentation procedures described in this protocol apply to all sponsor medical devices provided for use in the study. See Section 6.1.1 for the list of sponsor medical devices.

10.6.1. Definition of medical device AE and ADE

Medical device AE and ADE definition
• A medical device AE is any untoward medical occurrence, in a clinical study participant, users, or other persons, temporally associated with the use of study intervention whether or not considered related to the investigational medical device. An AE can therefore be any unfavorable and unintended sign (including an abnormal laboratory finding), symptom, or disease (new or exacerbated) temporally associated with the use of an investigational medical device. This definition includes events related to the investigational medical device or comparator and events related to the procedures involved except for events in users or other persons, which only include events related to investigational devices. • An ADE is defined as an AE related to the use of an investigational medical device. This definition includes any AE resulting from insufficient or inadequate instructions for use or any malfunction of the investigational medical device as well as any event resulting from use error or from intentional misuse of the investigational medical device.

10.6.2. Definition of medical device SAE, SADE and USADE

A Medical Device SAE is any serious AEs that:
a. Led to death
b. Led to serious deterioration in the health of the participant, that either resulted in: • A life-threatening illness or injury. The term ‘life-threatening’ in the definition of ‘serious’ refers to an event in which the participant was at risk of death at the time of the event. It does not refer to an event, which hypothetically might have caused death, if it were more severe. • A permanent impairment of a body structure or a body function. • Inpatient or prolonged hospitalization. Planned hospitalization for a pre-existing condition, or a procedure required by the protocol, without serious deterioration in health, is not considered an SAE.

• Medical or surgical intervention to prevent life-threatening illness or injury or permanent impairment to a body structure or a body function. • Chronic disease (MDR 2017/745).
c. Led to fetal distress, fetal death or a congenital abnormality or birth defect
SADE definition
• A SADE is defined as an ADE that has resulted in any of the consequences characteristic of an SAE. • Any device deficiency that might have led to an SAE if appropriate action had not been taken, intervention had not occurred, or circumstances had been less fortunate.
Unanticipated SADE (USADE) definition
• An USADE (also identified as UADE in US Regulations 21 CFR 813.3), is defined as a serious ADE that by its nature, incidence, severity or outcome has not been identified in the current version of the risk analysis report. (see Section 2.3).

10.6.3. Definition of device deficiency

Device deficiency definition
• A device deficiency is an inadequacy of a medical device with respect to its identity, quality, durability, reliability, safety, or performance. Device deficiencies include malfunctions, use errors, and inadequacy of the information supplied by the manufacturer.

10.6.4. Recording and follow-up of medical device AE and/or SAE and device deficiencies

10.6.4.1. Medical device AE, SAE, and device deficiency recording

- When an AE/SAE/device deficiency occurs, it is the responsibility of the investigator to review all documentation (e.g., hospital progress notes, laboratory reports, and diagnostics reports) related to the event.
- The investigator will then record all relevant AE/SAE/device deficiency information in the participant's medical records, in accordance with the investigator's normal clinical practice and on the appropriate form.
- It is not acceptable for the investigator to send photocopies of the participant's medical records to GSK in lieu of completion of the AE/SAE/device deficiency form.
- There may be instances when copies of medical records for certain cases are requested by GSK. In this case, all participant identifiers, with the exception of the participant number, will be redacted on the copies of the medical records before submission to GSK.

- The investigator will attempt to establish a diagnosis of the event based on signs, symptoms, and/or other clinical information. Whenever possible, the diagnosis (not the individual signs/symptoms) will be documented as the AE/SAE.
- For device deficiencies, it is very important that the investigator describes any corrective or remedial actions taken to prevent recurrence of the deficiency.
  - A remedial action is any action other than routine maintenance or servicing of a medical device where such action is necessary to prevent recurrence of a device deficiency. This includes any amendment to the device design to prevent recurrence.
- If the site during the course of the study becomes aware of any serious, non-serious incident (including device deficiencies and malfunctions) related to any GSK non-IMP product they will report these events to GSK or to the concerned CA via the national spontaneous reporting system. These will be classified as spontaneous ICSRs.

10.6.4.2. Assessment of intensity

Refer to Section 10.3.4.2

10.6.4.3. Assessment of causality

Refer to Section 10.3.4.3

10.6.4.4. Follow-up of medical device AE/SAE and device deficiency

- The investigator is obligated to perform or arrange for the conduct of supplemental measurements and/or evaluations as medically indicated or as requested by GSK to elucidate the nature and/or causality of the AE/SAE/device deficiency as fully as possible. This may include additional laboratory tests or investigations, histopathological examinations, or consultation with other health care professionals.
- If a participant dies during participation in the study or during a recognized follow-up period, the investigator will provide GSK with a copy of any post-mortem findings including histopathology.
- New or updated information will be recorded in the originally completed form.
- The investigator will submit any updated SAE data to GSK within 24 hours of receipt of the information.

10.6.5. Reporting of medical device SAEs

Medical Device SAE Reporting to GSK via an Electronic Data Collection Tool

- The primary mechanism for reporting an SAE to GSK will be the electronic data collection tool.
- If the electronic system is unavailable, then the site will use the paper SAE data collection tool (see next table) to report the event within 24 hours.

- The site will enter the SAE data into the electronic system as soon as it becomes available.
- After the study is completed at a given site, the electronic data collection tool will be taken offline to prevent the entry of new data or changes to existing data.
- If a site receives a report of a new SAE from a study participant or receives updated data on a previously reported SAE after the electronic data collection tool has been taken offline, then the site can report this information on a paper SAE form or to the medical monitor by telephone.
- If the site during the course of the study or poststudy becomes aware of any serious, non-serious AEs, pregnancy exposure, related to any GSK device they will report these events to GSK or to the concerned CA via the national spontaneous reporting system. These will be classified as spontaneous ICSRs.
- Contacts for SAE reporting can be found in Section 8.4.9.

Medical Device SAE Reporting to GSK via Paper Data Collection Tool

- Email/Facsimile transmission of the SAE paper data collection tool is the preferred method to transmit this information to the medical monitor.
- In rare circumstances and in the absence of email/facsimile equipment, notification by telephone is acceptable with a copy of the SAE paper data collection tool sent by overnight mail or courier service.
- Initial notification via telephone does not replace the need for the investigator to complete and sign the SAE paper data collection tool within the designated reporting time frames.
- Contacts for SAE reporting can be found in Section 8.4.9.

10.6.6. Reporting of SADEs

SADE Reporting to GSK

NOTE: There are additional reporting obligations for medical device deficiencies that are potentially related to SAEs that must fulfill the legal responsibility to notify appropriate regulatory authorities and other entities about certain safety information relating to medical devices being used in clinical studies.

- Any device deficiency that is associated with an SAE must be reported to the sponsor within 24 hours after the investigator determines that the event meets the definition of a device deficiency.
- The sponsor will review all device deficiencies and determine and document in writing whether they could have led to an SAE. These device deficiencies will be reported to the regulatory authorities and IRBs/IECs as required by national regulations.
- Contacts for SAE reporting can be found in Section 8.4.9.

10.6.7. Reporting of medical device deficiencies for associated person

• **Reporting to GSK**
If an Associated Person (i.e., e.g., spouse, caregiver, site staff) experiences a device deficiency, the medical device deficiency information, and any associated AE/SAE information will be reported to GSK. The associated person will be provided with the authorization to contact physician letter. If follow-up information is required, authorization to contact physician (or other licensed medical practitioner) must be signed to obtain consent. • Medical device deficiencies that are not related to an AE or SAE should be reported via email to gsk-rd.complaints@gsk.com, using the medical device deficiency report form. • If the medical device deficiency is related to a non-serious AE and not linked to an SAE, please send the medical device deficiency report form with details of the associated AE via email to gsk-rd.complaints@gsk.com only. • If the device incident is linked to an SAE, please email the medical device deficiency report form, within 24 hours. Refer to Section 8.4.9 for reporting. • GSK will review all device deficiencies and determine and document in writing whether they could have led to an SAE. These device deficiencies will be reported to the regulatory authorities and IRBs/IECs as required by national regulations.

10.7. Appendix 7: Standard Error (SE) Calculation for 2x2x4 Full Replicate Design

CCI

10.8. Appendix 8: Reference-Scaled Acceptance Range Limit

CCI

10.9. Appendix 9: Protocol amendment history

The Protocol Amendment Summary of Changes Table for the current amendment is located directly before the TOC.

Amendment [1]

Overall rationale for the Amendment

The objective of this amendment is to align the time period for collecting pregnancy information for female participants with the time period for post-intervention contraception. In addition, parameter specifications for microscopic evaluation have been removed.

11. REFERENCES

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