16 October 2025
EMA/345552/2025
Committee for Medicinal Products for Human Use (CHMP)

Assessment report

Brinsupri

International non-proprietary name: brensocatib

Procedure No. EMEA/H/C/005820/0000

Note

Assessment report as adopted by the CHMP with all information of a commercially confidential nature deleted.

Official address Domenico Scarlattilaan 6 ● 1083 HS Amsterdam ● The Netherlands
Address for visits and deliveries Refer to www.ema.europa.eu/how-to-find-us
Send us a question Go to www.ema.europa.eu/contact **Telephone** +31 (0)88 781 6000 An agency of the European Union

Table of contents

List of abbreviations

Abbreviation	Definition
ADR	adverse drug reaction
AE	adverse event
AESI	adverse event of special interest
ALT	alanine aminotransferase
ANC	absolute neutrophil counts
AST	aspartate aminotransferase
AUC	area under the concentration-time curve
AUC_{∞}	area under the plasma concentration-time curve from time zero to infinity
$AUC_{\square}$	area under the plasma concentration-time curve during a dosage interval (☐) at steady-state
BA	Bioavailability
BMI	body mass index
BSI	Bronchiectasis Severity Index
CatG	Cathepsin G
CHMP	Committee for Medicinal Products for Human Use
CL/F	total clearance after oral administration
CF	cystic fibrosis
C_{max}	maximum (peak) plasma concentration of the drug
C_{min}	minimum (trough) plasma drug concentration
CMQ	Customized MedDRA Query
CNS	central nervous system
COPD	chronic obstructive pulmonary disease
COVID-19	Coronavirus disease 2019
CPK	creatinine phosphokinase
CRSsNP	chronic rhinosinusitis without nasal polyps
CSR	clinical study report
CT	computed tomography
CV	coefficient of variation
CYP	cytochrome P450
DDI	drug-drug interaction

DPP1	dipeptidyl peptidase 1
EAP	expanded access program
EOS	end of study
FEV_1	forced expiratory volume in 1 second
GCP	Good Clinical Practice
IC_{50}	concentration of drug producing 50% inhibition
ICPD	Institute for Clinical Pharmacodynamics
ITT	intent-to-treat
MAD	multiple-ascending dose
MAR	missing at random
MATE	multidrug and toxin extrusion
NAG	N-acetyl-β-D-glucosaminidase
NCFBE	non-cystic fibrosis bronchiectasis
NE	neutrophil elastase
NOAEL	no-observed-adverse-effect level
NSP(s)	neutrophil serine protease(s)
PBPK	physiological based pharmacokinetic(s)
PD	pharmacodynamic(s)
PE	pulmonary exacerbation
Pgp	P-glycoprotein
PK	pharmacokinetic(s)
PLD	Phospholipidosis
PLS	Papillon-Lefèvre syndrome
$ppFEV_1$	percent predicted forced expiratory volume in 1 second
PPK	population pharmacokinetic(s)
PR3	proteinase 3
QD	once daily
QOL-B	Quality of Life Questionnaire-Bronchiectasis
RNA	ribonucleic acid
SAD	single ascending dose
SAE	serious adverse event
SOC	System Organ Class

SUSAR	serious unexpected suspected adverse reaction
$t_{½}$	elimination half-life
TEAE	treatment-emergent adverse event
ULN	upper limit of normal
US	United States
V_d/F	volume of distribution following extravascular dosing

1. Executive summary

On 16th October 2025, the Committee for Medicinal Products for Human Use (CHMP) adopted a positive opinion recommending the granting of a marketing authorisation for the medicinal product Brinsupri (brensocatib) intended for the treatment of non-cystic fibrosis bronchiectasis (NCFBE) in patients 12 years of age and older.

Brinsupri (brensocatib) will be available as a 25 mg oral, film-coated tablet. Brensocatib is a competitive, reversible inhibitor of dipeptidyl peptidase 1 (DPP1). Brensocatib reduces the activity of neutrophil serine proteases (NSPs) implicated in the pathogenesis of bronchiectasis, including neutrophil elastase, cathepsin G and proteinase 3.

The full indication for Brinsupri is treatment of non-cystic fibrosis bronchiectasis (NCFB) in patients 12 years of age and older with two or more exacerbations in the prior 12 months.

The main evidence of efficacy of Brinsupri was based on one Phase III randomised double-blind placebo controlled clinical trial (ASPEN) with a total of 1,721 patients aged 12 years and older. At 52 weeks, the annualised rate of exacerbations was 1.04 with brensocatib 25 mg versus 1.29 with placebo; the rate ratio was 0.81 (95% CI 0.69–0.94), demonstrating a statistically significant reduction versus placebo

The most commonly reported adverse reactions were headache (9.2%), hyperkeratosis (5.9%), gingival and periodontal diseases (5.3%), dermatitis (4.2%), rash (4.1%), upper respiratory tract infection (3.9%), and dry skin (3.0%). Use during pregnancy is not recommended; animal studies have shown reproductive (embryo-foetal) toxicity.

Detailed recommendations for the use of this product are described in the summary of product characteristics (SmPC), which will be published on the EMA website in all official European Union languages after the marketing authorisation has been granted by the European Commission.

This report summarises the scientific review leading to the opinion adopted by the Committee for Medicinal Products for Human Use (CHMP).

2. Administrative/regulatory information and recommendations on the procedure

2.1. Information on the product

Product data	
Product name	Brinsupri
Active substance	Brensocatib monohydrate
INN or common name	Brensocatib
Applicant	Insmed Netherlands B.V. Stadsplateau 7 3521 AZ Utrecht NETHERLANDS
EMA product number	EMEA/H/C/005820
ATC code and pharmacotherapeutic group	R03 - Drugs for obstructive airway disease
Pharmaceutical form(s) and strength (s)	Film-coated tablet 25 mg
Packaging	blister (PVC/PCTFE/alu)
Package size(s)	28 tablets
Route of administration	Oral use
Orphan designation	N/A
Orphan indication status confirmed	Not applicable
PRIME scheme	Granted on 27 Feb 2020
Type of marketing authorisation granted at opinion	Standard
Legal basis	Article 8.3 of Directive 2001/83/EC
Final indication	Brinsupri is indicated for the treatment of non-cystic fibrosis bronchiectasis (NCFB) in patients 12 years of age and older with two or more exacerbations in the prior 12 months.
New active substance status	Granted

2.2. Scientific advice

Please refer to the section on PRIME below.

2.3. PRIME

Brinsupri (brensocatib) was granted eligibility to PRIME (priority medicines) on 27 February 2020 in the following indication: treatment of non-cystic fibrosis bronchiectasis in patients 12 years of age and older with two or more exacerbations in the prior 12 months.

Eligibility to PRIME was granted at the time in view of the following:

- Unmet medical need for effective specifically authorised treatments of NCFBE is agreed
- The non-clinical data presented support a mechanism of action as a reversible DPP1 inhibitor which reduces the activation of neutrophile serin proteases (NSPs)
- The Phase 1 data show exposure dependent inhibition of Neutrophil Elastase (NE) activity in whole blood samples after 21/28d of treatment and in PK/PD analyses of the Ph2 study data, a dose proportional reduction of NE, cathepsin-G and Pr3 (all NSPs) in sputum over the study period has been shown, supporting DPP1 inhibition as important element of the mechanism of action
- Phase 2 data from a 3-arm double-blind, placebo controlled randomised trial in 256 subjects administering 10/25mg QD for 24 weeks support an assumption of efficacy in the target population: the primary endpoint of 'time-to-exacerbation' improved for both dose strengths, 'rate of exacerbations' improved with relevant effect size and subgroup analyses of the primary endpoint suggest efficacy across subgroups including severely affected patients with high exacerbation rates and in patients receiving macrolides
- Safety signals (clinical: skin, gingiva/periodontium; non-clinical: phospholipidosis kidney, lung; testicular tox) indicate need for careful safety characterisation to inform benefit/risk in the target population

Upon granting of eligibility to PRIME, the rapporteur was appointed by the CHMP.

The applicant was recommended to address the following key issues through relevant regulatory procedures:

Overview of selected previous CHMP scientific advice in the proposed indication in the past 5 years

SA procedure No. including year and file link	Product	Applicant	Indication
25/06/2020 EMEA/H/SA/4482/1/2020/III EMEA/H/SA/4482/1/2020/III	Brensocatib	Insmed Netherlands BV	Treatment of NCFBE
21/07/2022 EMA/SA/0000092783 EMA/SA/0000092783	Brensocatib	Insmed Netherlands BV	Treatment of NCFBE
26/04/2023 EMA/SA/0000126545 EMA/SA/0000126545	Brensocatib	Insmed Netherlands BV	Treatment of NCFBE
14/09/2023 EMA/SA/0000140961 EMA/SA/0000140961	Brensocatib	Insmed Netherlands BV	Treatment of NCFBE

09/11/2023 EMA/SA/0000149411 EMA/SA/0000149411	Brensocatib	Insmed Netherlands BV	Treatment of NCFBE
25/04/2024 EMA/SA/0000166717 EMA/SA/0000166717 Quality only advice	Brensocatib	Insmed Netherlands BV	Treatment of NCFBE

Scientific advice EMEA/H/SA/4482/1/2020/III pertained the questions about following non-clinical and clinical development issues:

Nonclinical Safety Profile: The nonclinical safety profile supports the initiation of a Phase 3 clinical program for INS1007.

- Additional Nonclinical Safety Studies: No further nonclinical safety studies are required beyond those outlined to support a marketing application.
- Clinical development, including patient population, sample size, dose selection, safety data, statistical methods and hypothesis.

SA outcome

- Single Phase 3 Study: A single Phase 3 study is adequate to support the efficacy and safety reviews for the proposed indication.
- Patient Population: The patient population - adults aged 18 to 85 years with a clinical history consistent with non-cystic fibrosis bronchiectasis (NCFBE) with at least two documented pulmonary exacerbations in the past 12 months, planned for the study is appropriate to support the intended indication.
- Dose Selection: The dose selection of 10 mg and 25 mg for Phase 3 is appropriate.
- Safety Assessments: The proposed safety assessments and monitoring plan for the Phase 3 study are acceptable.
- Safety Database: The proposed safety database is adequate to characterize the safety profile of INS1007 to support a marketing application.
- Study Sample Size: The proposed study sample size, planned interim analysis, and sample size reassessment procedure are acceptable.
- Statistical Hypothesis Testing and methods: The proposed statistical hypothesis testing procedures to handle multiplicity for controlling study-wise type I error rate are acceptable.
- Clinical Pharmacology Program: The overall clinical pharmacology program is adequate to support regulatory submission.

Scientific Advice EMA/SA/0000092783 pertained the questions about following clinical development issues:

EPAR summary

- Adequacy of the proposed statistical analysis plan for the ongoing single pivotal Phase 3 study INS1007-301, including estimands and analytical methods for primary and secondary endpoints; proposal to exclude all data from patients located in Ukraine due to the ongoing war.

SA outcome

The scientific advice addressed several clinical aspects, including the adequacy of the statistical analysis plan for the ongoing pivotal Phase 3 study INS1007-301, particularly concerning estimands and handling of missing data.

CHMP noted that major changes to the study design should be avoided, and the estimand framework should clarify the statistical analysis questions. They discussed intercurrent events affecting the primary endpoint and suggested separate strategies for handling these events.

The CHMP expressed concerns regarding the missing data assumptions and recommended sensitivity analyses to assess the robustness of the primary analysis results. They emphasized the need for a clear justification for the missing data handling approach.

Exclusion of Data from Ukraine - Due to the ongoing war in Ukraine, the applicant proposed to exclude data from patients in Ukraine from efficacy and safety analyses, which the CHMP found acceptable given the circumstances. They recommended performing sensitivity analyses when possible.

Scientific Advice EMA/SA/0000126545 pertained the questions about following clinical development issues:

EPAR summary

- Adequacy of the characterisation of the metabolic profile for one major and other non-major metabolites; sufficiency of the proposed plan for PPK and exposure response analyses; dosing optimization in particular subgroups of populations and the need of an additional DDI study with rifampin and esomeprazole to complete the interactions-profile of brensocatib.

SA outcome

Metabolic Profile: The CHMP concluded that while the data presented by the applicant seem to support that there is no need to further characterize the metabolic profile for M8 and other non-major metabolites, a definite answer will only be possible after thorough assessment of comprehensive data at the time of MAA.

PPK and Exposure Response Analyses: The CHMP agreed that the proposed plan for PPK and exposure-response analyses is acceptable in principle, provided that all relevant categories/covariates are adequately represented in the analysis dataset. However, it is considered late to plan dose optimization in the entire target population.

DDI Study: The CHMP agreed to the plan by the applicant to conduct an additional DDI study with rifampin and esomeprazole to complete the interaction profile of brensocatib.

Agreement with the applicant:

The CHMP supports the rationale for the need of an additional DDI study with rifampin and esomeprazole.

The final decision on the need for additional in vivo DDI studies will only be possible once a complete data package has been submitted.

Main Caveats:

The adequacy of the metabolic profile characterization will be assessed at the time of MAA.

The interpretation of the current exposure-response analysis results is not fully understood.

It is considered late to plan dose optimization in the entire target population because of ongoing study.

Scientific Advice EMA/SA/0000140961 pertained the questions about following multidisciplinary quality and clinical development issues:

EPAR summary

- The need for dedicated relative bioavailability studies to bridge between phase 2, phase 3 and to-be-marketed formulations.

The advice covered quality and clinical aspects, including the adequacy of the formulation bridging strategy between Phase 2 and Phase 3 tablets, dissolution testing results, and the need for additional in vitro studies. The CHMP emphasized the importance of complete clinical data and PK modeling results but noted that the final decision on the waiver for dedicated bioavailability studies would be made during the marketing authorization assessment.

Scientific Advice EMA/SA/0000149411 pertained the questions about following development issues:

EPAR summary

- Study data cut-off for the statistical analyses in an ongoing Phase 3, randomised, double-blind, placebo-controlled, parallel-group, multi-centre study to assess the efficacy, safety, and tolerability of brensocatib administered once daily for 52 weeks in subjects with non-cystic fibrosis bronchiectasis; age group (adults vs adolescents) as a categorical covariate in the primary analysis model and additional subgroup analyses by age group in the same study.

SA outcome

The SA includes questions on the clinical development of brensocatib, focusing on the study data cut-off for statistical analyses, randomisation ratios, and the inclusion of age groups in the primary analysis model. Specifically, it addresses the adequacy of including age group as a categorical covariate in the primary analysis model and additional subgroup analyses by age group. The questions also cover the approach for the INS1007-301 study data cut for the statistical analyses for the proposed indication in the upcoming Marketing Authorisation Application (MAA), and whether the inclusion of age group (adults vs adolescents) as a categorical covariate in the primary analysis model and additional subgroup analyses by age group will be adequate and sufficient to address concerns about bias introduced by different randomisation ratios and possible differences in treatment effects between adults and adolescents.

The CHMP agreed on the approach for the INS1007-301 study data cut for the statistical analyses for the proposed indication in the upcoming MAA. The data cut defined for the primary analysis would be acceptable, but it does not endorse the design with different randomisation ratios for adults and adolescents and simple pooled analysis. The CHMP emphasized that the agreed Paediatric Investigation Plan (PIP) specifies the inclusion of at least 40 adolescents evaluable for the primary analysis.

The CHMP does not consider using different randomisation ratios for adults and adolescents in the same study and implementing a primary analysis that evaluates the two cohorts together by simple pooling as an appropriate approach. The applicant should carefully consider the options outlined, and a primary analysis without adolescents could be preferred. The CHMP advised that extrapolation of efficacy and safety from the adult data will also depend on PK modelling and simulation data.

2.4. Eligibility to the centralised procedure

The applicant Insmed Netherlands B.V. submitted on 26 March 2025 an application for marketing authorisation to the European Medicines Agency (EMA) for Brinsupri (Brensocatib), through the centralised procedure under Article 3 (2) (a) of Regulation (EC) No 726/2004. The eligibility to the centralised procedure was agreed upon by the EMA/CHMP on 12 November 2020.

The applicant applied for the following indication: treatment of non-cystic fibrosis bronchiectasis in patients 12 years of age and older with two or more exacerbations in the prior 12 months.

2.5. Legal basis and dossier content

The legal basis for this application refers to:

Article 8.3 of Directive 2001/83/EC - complete and independent application

The application submitted is composed of administrative information, complete quality data, and non-clinical and clinical data based on applicant's own tests and studies and bibliographic literature substituting/supporting certain test(s) or studies.

2.6. Information on paediatric requirements

Pursuant to Article 8 of Regulation (EC) No 1901/2006, the application included an EMA decision(s) P/0403/2021 on the agreement of a paediatric investigation plan (PIP).

At the time of submission of the application, the PIP P/0403/2021 had not yet been completed as some measures had been deferred.

2.7. Information on orphan market exclusivity

2.7.1. Similarity with authorised orphan medicinal products

Pursuant to Article 8 of Regulation (EC) No 141/2000 and Article 3 of Commission Regulation (EC) No 847/2000, the applicant did not submit a critical report addressing the possible similarity with authorised orphan medicinal products from the start of the procedure because there is no authorised orphan medicinal product for a condition related to the proposed indication.

2.8. Applicant's request(s) for consideration

2.8.1. Accelerated assessment request

The applicant requested accelerated assessment in accordance to Article 14 (9) of Regulation (EC) No 726/2004. The CHMP agreed to the applicant's request for an accelerated assessment as the product was considered to be of major public health interest. This was based on the following:

Brensocatib, a first in class DPP1 inhibitor, targets chronic inflammation by inhibiting neutrophil serine proteases (NSPs), breaking the cycle of airway damage and infection, and constitutes a therapeutic innovation. Top line results from the Phase III ASPEN study demonstrated brensocatib's efficacy in reducing moderate/severe PEs and prolonging exacerbation-free periods. Brensocatib showed a favourable safety profile, with treatment emergent advice events (TEAEs) comparable to placebo. It exhibited no significant drug interactions or pharmacokinetic alterations in renal or hepatic impairment, supporting its potential as a safe, once-daily treatment for NCFBE. Presented clinical data seems robust and clinically important and are expected to enable in depth assessment of the benefit and risk.

2.8.2. New active substance status

The applicant requested the active substance brensocatib monohydrate contained in the above medicinal product to be considered as a new active substance, as the applicant claims that it is not a constituent of a medicinal product previously authorised within the European Union.

2.8.2.1. CHMP recommendation on new active substance status

Based on the review of available data on the active substance, the CHMP considers that brensocatib is to be qualified as a new active substance in itself, as it is not a constituent of a medicinal product previously authorised within the European Union.

2.9. Steps taken for the assessment of the product

The Rapporteur and Co-rapporteur appointed by the CHMP were:

Rapporteur:	Janet Koenig
Co-rapporteur:	Selma Arapovic Dzakula

The application was received by the EMA on	26 March 2025
An application for accelerated assessment was filed by the applicant	27 February 2025
The procedure started on	24 April 2025
The CHMP rapporteur's first assessment report was received on	24 June 2025
The CHMP Co-rapporteur's first assessment report was added to the rapporteur's report on	26 June 2025
The PRAC rapporteur's first assessment report was added to the rapporteurs' report and circulated to all PRAC and CHMP members on	01 July 2025
In accordance with Article 6(3) of Regulation (EC) No 726/2004, the CHMP rapporteur and Co-rapporteur declared that they had completed their assessment report in less than 80 days	
The PRAC agreed on the PRAC Assessment Overview and Advice to CHMP during the meeting on	10 July 2025
The CHMP agreed on the consolidated list of questions (LoQ) to be sent to the applicant during the meeting on	22 July 2025
The applicant submitted the responses to the CHMP consolidated List of Questions on	13 August 2025
The CHMP rapporteur circulated the CHMP and PRAC rapporteurs joint assessment report on the applicant's responses to the list of questions (LoQ) to all CHMP and PRAC members on	05 September 2025
The CHMP rapporteur circulated the CHMP and PRAC rapporteurs joint updated assessment report on the applicant's responses to the list of questions (LoQ) to all CHMP and PRAC members on	11 September 2025
The CHMP agreed on a list of outstanding issues (LoOI) to be sent to the applicant on	16 September 2025
The applicant submitted the responses to the CHMP list of outstanding issues on	22 September 2025
The CHMP rapporteur circulated the CHMP and PRAC rapporteurs Joint assessment report on the applicant's responses to the list of outstanding issues to all CHMP and PRAC members on	02 October 2025
The CHMP rapporteur circulated the CHMP and PRAC rapporteurs Joint updated assessment report on the applicant's responses to the list of outstanding issues to all CHMP and PRAC members on	09 October 2025
The CHMP, in the light of the overall data submitted and the scientific discussion within the Committee, issued a positive opinion for granting a marketing	16 October 2025

authorisation to Brinsupri on	
Furthermore, the CHMP adopted a report on New Active Substance (NAS) status of the active substance contained in the medicinal product.	16 October 2025

2.10. Final CHMP outcome

2.10.1. Considerations related to paediatrics

The CHMP reviewed the available paediatric data of studies subject to the agreed paediatric investigation plan (PIP) P/0403/2021 and the results of these studies are reflected in the summary of product characteristics (SmPC) and, as appropriate, the package leaflet. Relevant paediatric statement in Section 5.1 of the SmPC if the EMA has deferred a paediatric development have also been included.

2.10.2. Final opinion

Based on the CHMP review of data on quality, safety and efficacy, the CHMP considers by consensus that the benefit-risk balance of Brinsupri is favourable in the following indication:

Brinsupri is indicated for the treatment of non-cystic fibrosis bronchiectasis (NCFB) in patients 12 years of age and older with two or more exacerbations in the prior 12 months.

The CHMP, therefore recommends the granting of the marketing authorisation subject to the conditions described in the following sections.

Furthermore, following the review of the available data in the context of the applicant's claim of new active substance status, the CHMP position is reflected in a separate report (not published).

2.10.3. Conditions or restrictions regarding supply and use

Medicinal product subject to medical prescription.

2.10.4. Other conditions and requirements of the marketing authorisation

2.10.4.1. Periodic safety update reports

The requirements for submission of periodic safety update reports for this medicinal product are set out in the list of Union reference dates (EURD list) provided for under Article 107c(7) of Directive 2001/83/EC and any subsequent updates published on the European medicines web-portal.

2.10.5. Conditions or restrictions with regard to the safe and effective use of the medicinal product

2.10.5.1. Risk management plan (RMP)

The marketing authorisation holder (MAH) shall perform the required pharmacovigilance activities and interventions detailed in the agreed RMP presented in Module 1.8.2 of the marketing authorisation and any agreed subsequent updates of the RMP.

An updated RMP should be submitted:

- At the request of the European Medicines Agency;
- Whenever the risk management system is modified, especially as the result of new information being received that may lead to a significant change to the benefit/risk profile or as the result of an important (pharmacovigilance or risk minimisation) milestone being reached.

2.10.5.2. Obligation to conduct post-authorisation measures

The MAH shall complete, within the stated timeframe, the below measures:

Table 1: Post-authorisation measures

Description	*Due date*
Non- interventional post authorisation safety study (PASS): Evaluation of the long term safety in patients treated with Brinsupri in the real world setting	Q4 2034

2.10.6. Conditions or restrictions with regard to the safe and effective use of the medicinal product to be implemented by the Member States

Not applicable.

2.10.7. Proposed list of recommendations

Table 2: Proposed list of recommendations

Description of recommendation(s)
1. The applicant is recommended to include an additional point in the validation of linearity for the determination method of an elemental impurity (REC1).
2. The applicant is recommended to provide stability data on two additional stability batches (REC2).
3.A post authorisation *in vivo* CYP3A4 induction study: to evaluate the effect of steady-state brensocatib on the pharmacokinetics of a sensitive CYP3A4 substrate, midazolam and 1-OH-midazolam in healthy volunteers

3. Introduction

3.1. Therapeutic context

Brensocatib (previously INS1007, AZD7986) is a first-in-class, oral, selective, competitive, and reversible inhibitor of dipeptidyl peptidase 1 (DPP1). DPP1 plays a critical role in the activation of neutrophil serine proteases (NSPs)—including neutrophil elastase (NE), proteinase 3 (PR3), and cathepsin G (CatG)—during neutrophil maturation in the bone marrow. These NSPs are implicated in the pathogenesis of multiple neutrophil-driven inflammatory diseases, including non-cystic fibrosis bronchiectasis (NCFBE).

By inhibiting DPP1, brensocatib prevents the activation of these NSPs, thereby reducing their harmful activity in tissues such as the lung. This mechanism is particularly relevant to NCFBE, where excessive NSP activity leads to tissue damage, mucus hypersecretion, and sustained inflammation, contributing to disease progression and pulmonary exacerbations. Brensocatib's mechanism of action is supported by pharmacodynamic data showing dose- and exposure-dependent reductions in active NSP levels in sputum, blood, and plasma.

The inhibition of DPP1 offers a novel therapeutic approach to modulate neutrophilic inflammation at its source, distinguishing brensocatib from existing symptomatic and anti-infective therapies in bronchiectasis, none of which specifically target NSP activity.

3.2. Aspects of development

The clinical development program for brensocatib was designed to evaluate its safety, efficacy, pharmacokinetics (PK), and pharmacodynamics (PD) in the treatment of non-cystic fibrosis bronchiectasis (NCFBE). The program includes 13 completed clinical studies and ongoing studies, including an expanded access program (EAP).

Key completed studies comprise:

- Ten Phase 1 studies in healthy volunteers, which established the PK/PD profile, food effect, drug-drug interaction (DDI) potential, and tolerability of brensocatib.
- Phase 2 study in NCFBE (INS1007-201), which provided initial efficacy and safety data and informed dose selection.
- Phase 3 pivotal study in NCFBE (INS1007-301), designed to confirm the efficacy and safety of brensocatib 10 mg and 25 mg once daily (QD) over 52 weeks.
- Phase 2 study in cystic fibrosis (INS1007-211) and supportive PK and safety data from additional studies, including one in chronic rhinosinusitis without nasal polyps (CRSsNP).

The applicant considers the results of the Phase 2 and Phase 3 studies to support the proposed use of brensocatib 25 mg once daily in patients aged 12 years and older with NCFBE and a recent history of exacerbations.

3.3. Description of the product

Brensocatib is being developed as an immediate-release, film-coated oral tablet, available in two strengths: 10 mg and 25 mg. The proposed commercial presentation is a grey, debossed tablet, consistent in core formulation, size, and shape with the tablet used in the Phase 3 clinical study, differing only in film-coating colour from the clinical trial batches. The 25 mg tablet is the applicant's applied for.

As of the clinical data cut-off date (28 March 2024), brensocatib has not received marketing authorization for any indication from regulatory agencies. A marketing authorisation application (MAA) has been submitted in the European Union for the treatment of NCFBE in patients aged 12 years and older with a history of ≥2 pulmonary exacerbations in the past 12 months.

The applicant has received scientific advice and protocol assistance from the European Medicines Agency (EMA) and other regulatory authorities throughout the development process. These interactions addressed the design of the pivotal Phase 3 study (INS1007-301), statistical methodologies, dose selection, inclusion of adolescent participants, and the overall clinical pharmacology strategy.

3.4. Inspection issues

3.4.1. Good manufacturing practice (GMP) inspection(s)

No inspection required.

3.4.2. Good laboratory practice (GLP) inspection(s)

No inspection required.

3.4.3. Good clinical practice (GCP) inspection(s)

Based on the review of clinical data, CHMP did not identify the need for a GCP inspection of the clinical trials included in this dossier.

4. Quality aspects

4.1. Introduction

The finished product is presented as film-coated tablets containing 25 mg of brensocatib (as monohydrate) as active substance.

Other ingredients are:

Tablet core: cellulose, microcrystalline; calcium hydrogen phosphate dihydrate; sodium starch glycolate; silica, colloidal hydrated; glycerol dibehenate.

Film-coating: poly(vinyl alcohol); titanium dioxide (E171); macrogol; talc; black iron oxide (E172).

The product is available in PVC/PCTFE aluminium foil blister card containing 14 film-coated tablets.

4.2. Active substance

4.2.1. General information

The active substance is brensocatib monohydrate. The chemical name of brensocatib monohydrate is (2S)-N-{(1S)-1-cyano-2-[4-(3-methyl-2-oxo-2,3-dihydro-1,3-benzoxazol-5-yl)phenyl]ethyl}- 1,4-oxazepane-2-carboxamide monohydrate corresponding to the molecular formula $C_{23}H_{24}N_4O_4 \bullet H_2O$. It has a molecular mass of 438.48 g/mol and the following structure, depicted in Figure 1:

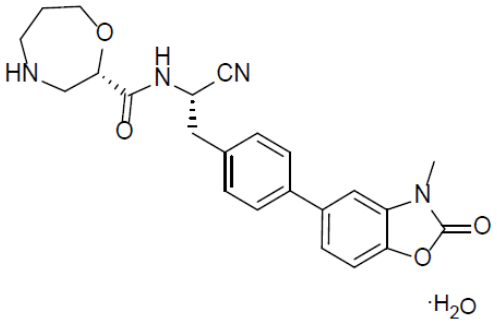


Figure 1: active substance structure

The chemical structure of brensocatib was elucidated by a combination of mass spectrometry (MS), nuclear magnetic resonance spectroscopy (NMR), proton (1H) NMR, carbon (13C) NMR, infrared transmittance spectrum (IR), ultraviolet spectrum (UV). The solid state properties of the active substance were measured by differential scanning calorimetry (DSC), thermal gravimetric analysis (TGA) and by X-ray powder diffraction (XRPD).

Brensocatib exhibits stereoisomerism due to the presence of two chiral centres.

Polymorphism has been observed for brensocatib. The most stable form was chosen for the development of the finished product.

Brensocatib is a a white to off-white solid powder, non-hygroscopic.

4.2.2. Manufacture, characterisation, and process controls

Brensocatib active substance is prepared using well defined regulatory starting materials (RSM) with acceptable specifications.

Adequate in-process controls are applied during the synthesis. The specifications and control methods for intermediate products, starting materials and reagents have been presented.

The characterisation of the active substance and its impurities are in accordance with the EU guideline on chemistry of new active substances.

Potential and actual impurities were well discussed with regards to their origin and characterised.

The removal of residual solvents is demonstrated and the risk of their carry over to the final active substance is considered negligible.

The commercial manufacturing process for the active substance was developed in parallel with the clinical development program. Changes introduced have been presented in sufficient detail and have been justified. . .The quality of the active substance used in the various phases of the development is considered to be comparable with that produced by the proposed commercial process. Stability and PPQ batches were produced in line with the proposed commercial process.

No process validation is needed, since the active substance is not sterile.

The primary packaging material is in line with Ph. Eur. 3.1.3 and EC Commission Regulation (EU)10/2011, as amended.

4.2.3. Specification

The active substance release and stability specification includes tests for: appearance, identification (IR, HPLC), assay (UPLC), impurities (UPLC), chiral impurities (HPLC), residual solvents (GC), water content (KF), elemental impurity content (ICP-MS) and sulfated ash (Ph. Eur.).

For the maximum daily dose (MDD) of 25 mg/day, a reporting threshold of 0.05%, an identification threshold of 0.10% and a qualification threshold of 0.15% for impurities, are applicable. Specified impurities are qualified and their limits can be accepted.

The analytical methods used have been adequately described and non-compendial methods appropriately validated in accordance with the ICH guidelines. Satisfactory information regarding the reference standards used for assay and impurities testing has been presented.

Batch analysis data on full scale commercial batches of the active substance are provided. The results are within the specifications and consistent from batch to batch.

Stability

Stability data from three commercial scale batches of active substance from the proposed manufacturer stored in a container closure system representative of that intended for the market under long term conditions and for up to 6 months under accelerated conditions according to the ICH guidelines were provided. The parameters tested are appearance, assay (UPLC), impurities (UPLC) and water content (KF) All tested parameters were within the specifications.

Photostability testing following the ICH guideline Q1B was performed on samples of the active substance. TResults on stress conditions thermal, acidic, basic, neutral solution, oxidative and high humidity were also provided on samples of the active substance.

The stability results indicate that the active substance manufactured by the proposed supplier is sufficiently stable. The stability results justify the proposed retest period. The post approval stability protocol is appropriate.

4.3. Finished medicinal product

4.3.1. Description of the product and pharmaceutical development

Brensocatib film-coated tablets are an immediate-release oral dosage form containing 25 mg of the active substance brensocatib (as monohydrate, equivalent to 26.07 mg of brensocatib monohydrate). Brensocatib film-coated tablets are round, gray tablets debossed with "25" on one side and "BRE" on the other side. The tablet dimensionsare stated in 3.2.P.1.

All excipients are compliant with Ph. Eur. standards, except for black iron oxide (E172) of the film-coating, where reference is made to the NF. For the film coat system, all components are given with respective quantities. Compliance of the black iron oxide and all other components of the film-coating system with Regulation (EU) 231/2012 for food additives has been confirmed. There are no novel excipients used in the finished product formulation. The list of excipients is included in section 6.1 of the SmPC and in paragraph 4.1 of this report.

No overages are used during manufacture of brensocatib film-coated tablets.

The pharmaceutical development of the finished product contains QbD elements.

The quality target product profile (QTPP) and related critical quality attributes (CQAs) have been identified.

The key properties of the active substance brensocatib monohydrate have been discussed in relation to the pharmaceutical development.

The formulation development is supported by an adequate discussion of the excipient selection; the function and the chosen grade of each excipient has been adequately presented. The relevant functionality-related characteristics of each excipient are controlled in the respective excipient specifications.

Compatibility of the excipients and the active substance is supported by stability study results.

The choice of the QC dissolution method has been justified. During the procedure a major objection (MO1) was raised in relation to the acceptability of the dissolution method questioning the following: 1. its discriminatory power; 2. the proposed acceptance criterion for dissolution in the release and the shelf-life specification; 3. results obtained ata specified pH; and 4. requesting the batch size of the batches used in comparative dissolution testing In response to MO1, the applicant did not conduct new studies to demonstrate the discriminatory power of the dissolution method. The argumention provided was accepted in line with guidance. A tightened dissolution limit was accepted in line with guidance based on dissolution data obtained from the clinical batches, which was endorsed. The *in vitro* dissolution behaviour at specified pH was not further pursued as the *in vivo* relevance of the unexpected findings was deemed negligible, as discussed in the clinical assessmentBatch sizes of the batches used during development and for formulation bridging were supplemented and considered adequate.

The suitability of the tablets (e.g. choice of excipients, swallowability, palatability), for the paediatric population (12 years of age and older) has been provided and it is satisfactory.

The manufacturing of the film coated tablet formulation is a conventional manufacturing process. The

manufacturing development has been evaluated through the use of risk assessment, design of experiments and other modelling techniques to identify the critical process parameters. A risk analysis was performed using the failure mode effect analysis (FMEA) method in order to define critical process steps and process parameters that may have an influence on the finished product quality attributes. The risk identification was based on the prior knowledge of products with similar formulations and manufacturing processes as well as on the experience from formulation development, process design and scale-up studies. The critical process parameters have been adequately identified

The primary packaging selected for commercial use of brensocatib film-coated tablet is a PVC (polyvinyl chloride)/PCTFE (Polychlorotrifluoroethylene) formable laminated blister with push through aluminium lidding, which is common for the chosen pharmaceutical form. The material complies with Ph.Eur. and EC requirements. The choice of the container closure system has been validated by stability data and is adequate for the intended use of the product.

4.3.2. Manufacture of the product and process controls

Satisfactory GMP documentation has been provided.

The process is considered to be a standard manufacturing process. The description of the manufacturing process is adequately detailed. A flow scheme of the manufacturing process is provided.

Prior to the packaging of the film-coated bulk tablets in the commercial blister packaging configuration, a holding time in the bulk container at predefined conditions has been proposed and is supported by data. Major steps of the manufacturing process have been validated by a number of studies. It has been demonstrated that the manufacturing process is capable of producing the finished product of intended quality in a reproducible manner. The in-process controls are adequate for this manufacturing process.

4.3.3. Product specification

The finished product release and end of shelf-life specification include appropriate tests for this kind of dosage form:appearance (visual), assay (UPLC), identification (UV and UPLC), content uniformity (Ph. Eur.), degradation products (UPLC), dissolution (Ph. Eur.), water content (Ph. Eur.), specified impurities (UPLC), hardness (Ph. Eur.) and friability (Ph. Eur.). The finished product is released on the market based on the release specifications, through traditional final product release testing.

The maximum daily dose of brensocatib is 25 mg. Hence, the identification threshold for individual unspecified impurities is 0.20% and the qualification threshold for identified, specified impurities is of 0.50%. Specified impurities are limited either with the Q3B qualification threshold or with lower limits according to actual batch data. The limit for total impurities and water content were tightened tin line with batch data.

Following identification of an unspecified impurity, which caused OOS results in the accelerated stability studies, the impurity was added, as specified, to the finished product specification.

Omission of polymorph testing from the finished product specification at realase and end of shelf life has been justified with data.

Omission of microbial testing is considered acceptable. The analytical methods used have been adequately described and appropriately validated in accordance with the ICH guidelines.

The potential presence of elemental impurities in the finished product has been assessed following a risk-based approach in line with the ICH Q3D Guideline for Elemental Impurities. Batch analysis data

on three batches, tested with a validated method, were provided, demonstrating that each relevant elemental impurity was not detected above 30% of the respective PDE. The risk assessment was updated during the procedure to include information on the proposed commercial container closure system. Based on the updated risk assessment and the presented batch data it can be concluded that it is not necessary to include any elemental impurity controls in the finished product. An elemental impurity is adequately controlled. However, the applicant is recommended to include an additional point in the validation of linearity for its determination method (REC1). The information on the control of elemental impurities is satisfactory.

A risk assessment concerning the potential presence of nitrosamine impurities in the finished product has been performed considering all suspected and actual root causes in line with the "Questions and answers for marketing authorisation holders/applicants on the CHMP Opinion for the Article 5(3) of Regulation (EC) No 726/2004 referral on nitrosamine impurities in human medicinal products" (EMA/409815/2020) and the "Assessment report- Procedure under Article 5(3) of Regulation EC (No) 726/2004- Nitrosamine impurities in human medicinal products" (EMA/369136/2020). During the procedure, the proposed limit for a nitrosamine active substance-related impurity was deemed acceptable by the Nitrosamines Safety Operational Expert Group (NSOEG) as the outcome of the performed enhanced Ames Test was judged to be negative, based on a weight-of-evidence approach. Reference is made to the non-clinical assessment for further details.

The major root cause for the formation of a nitroso impurity has been identified, addressed and mitigated.

Absence of control of other impurity-related nitrosamines in the finished drug product was justified.

Based on the information provided, the risk of nitrosamine impurities in the active substance or the related finished product has been adequately addressed. Therefore, no specific additional control measures are deemed necessary.

Batch analysis results are provided for three commercial scale PPQ batches confirming the consistency of the manufacturing process and its ability to manufacture to the intended product specification.

Satisfactory information regarding the reference standards used for assay and impurities testing has been presented. In addition, a certificate of analysis for the reference standard of the nitroso impurity was submitted. The reference standard has been characterised by IR spectroscopy and $^{1}H/^{13}C$-NMR.

4.3.4. Stability of the product

Stability data from six commercial scale batches of finished product stored for up to 18 months under long term conditions, and for up to six months under accelerated conditions according to the ICH guidelines were provided for batches manufactured and were packed in the primary packaging proposed for marketing.

Stability-indicating parameters chosen for the stability protocol are appropriate.

The stability commitment in section P.8.2 has been amended with a statement that appropriate action will be taken in case of OOS results.

the applicant is recommended to further provide stability data on two additional stability batchesPhotostability studies in accordance with the conditions outlined in ICH Q1B were conducted. Based on the results, the finished product is considered to not be susceptible to light-induced degradation. Therefore, no additional storage statement for light protection is required.

Forced degradation studies have been conducted including an evaluation of the mass balance of the

methods to demonstrate the stability-indicating nature of the assay and degradation product method. Initially raised concerns and issues regarding the lack of mass balance at oxidative and base stress conditions have been satisfactorily cleared up by the applicant. In addition, provided verification of the microbiological purity method in accordance with Ph. Eur. has been added to the dossier for completeness.

Results of temperature cycling studies as mentioned by the applicant to corroborate the product's resistance to temperature excursions are presented and are acceptable.

The analytical procedures used are stability indicating as confirmed by the forced degradation studies. Based on available stability data, the proposed shelf-life of 18 months and 'Store in the original package in order to protect from moisture' with no storage temperature restriction, as stated in the SmPC (section 6.3 and 6.4) are acceptable.

4.3.5. Post approval changes management protocol

Not applicable.

4.3.6. Adventitious agents

Not applicable.

4.4. Discussion and conclusions on chemical, pharmaceutical and biological aspects

Information on development, manufacture and control of the active substance and finished product has been presented in a satisfactory manner.

During the procedure a MO was raised in relation to the adequacy of the QC dissolution method. The MO was resolvedby tightening the specification limits, by providing additional data on the batches tested and by demonstrating that the discriminatory power meets the guidance requirements.

During the procedure, points related to a nitrosamine impurity are considered resolved following the outcome of the NSOEG consultation and the submission of additional stability data.

The results of tests carried out indicate consistency and uniformity of important product quality characteristics, and these in turn lead to the conclusion that the product should have a satisfactory and uniform performance in clinical use.

At the time of the CHMP opinion, there were a number of minor unresolved quality issues having no impact on the Benefit/Risk ratio of the product, which pertain to the validation of an elemental impurity method and the provision of additional stability data. These points are put forward and agreed as recommendations for future quality development.

4.4.1. Conclusions on the chemical, pharmaceutical and biological aspects

The quality of this product is considered to be acceptable when used in accordance with the conditions defined in the SmPC. Physicochemical and biological aspects relevant to the uniform clinical performance of the product have been investigated and are controlled in a satisfactory way.

4.5. Recommendation for future quality development

In the context of the obligation of the MAHs to take due account of technical and scientific progress, the CHMP recommends the following points for investigation:

1. The applicant is recommended to include an additional point in the validation of linearity for the determination method for determination of an elemental impurity (REC1). (REC1).

2. The applicant is recommended to provide stability data on two additional stability batches (REC2).

5. Non-clinical aspects

5.1. Introduction

Brensocatib (INS1007, AZD7986 and AZ13661057) is a selective, competitive, and reversible inhibitor of the lysosomal cysteine proteinase dipeptidyl peptidase 1 (DPP1). It is being developed for chronic administration.

5.2. Analytical methods

Fully validated LC-MS/MS analytical assays for the quantification of brensocatib were used in the pivotal (GLP-compliant) toxicology studies.

Validated LC-MS/MS bioanalytical methods fit-for-purpose were used in PK and in vitro assay. support non-GLP and GLP studies as appropriate.

5.3. Pharmacology

5.3.1. Pharmacodynamics

5.3.1.1. Primary pharmacodynamics

Brensocatib is a first-in-class, oral, selective, competitive, and reversible inhibitor of dipeptidyl peptidase 1 (DPP1).

DPP1, also known as cathepsin C (CatC) is a highly conserved lysosomal cysteine dipeptidyl aminopeptidase belonging to the papain family. The best characterized physiological function of CatC is the activation of pro-inflammatory granule-associated serine proteases, including neutrophil elastase-like proteases (NPS), including neutrophil elastase (NE), proteinase 3 (PR3), and cathepsin G (CatG) during neutrophil maturation in the bone marrow. These proteases are synthesized as inactive zymogens containing a N-terminal pro dipeptide which maintains the zymogen in its inactive conformation and prevents premature activation potentially toxic to the cell. The activation occurs through cleavage of the N-terminal dipeptide by CatC during neutrophil maturation in the bone marrow.

The proposed indication of brensocatib is the treatment of non-cystic fibrosis bronchiectasis (NCFBE). NCFBE is a severe lung disease characterized by localized, irreversible enlargement of bronchi and bronchioles that may lead to obstructed breathing caused by abnormal mucus production. Activation of neutrophils in the airway leads to release of neutrophil serine proteases (NSPs) including neutrophil elastase (NE), proteinase 3 (PR3), and cathepsin G (CatG), which are believed to be central to

bronchiectasis pathophysiology (Chalmers and Chotirmall, 2018). Elevated NE, PR3 and CatG overwhelm natural inhibitors, such as alpha-1 antitrypsin and secretory leukoproteinase inhibitor, which leads to damaged airway walls (Chalmers et al., 2017), mucus hypersecretion, exacerbated inflammation, and disabled neutrophil and macrophage functions, increasing the risk of infection (Palmér et al., 2018). Thus, a reduction in NSP activities in response to brensocatib treatment provides an indirect measure of nonclinical efficacy.

In Vitro

Proposed Mode of Action of brensocatib:

Brensocatib is a selective, reversible inhibitor of didpetidyl peptidase I (DPP1). DPP1 is an enzyme that is responsible for the activation of NSPs in the bone marrow during the maturation of neutrophils. Brensocatib inhibits DPP1 and thus the activation of NSPs (including NE, PR3, and CatG). NSPs are involved in pathogen destruction and inflammation mediation, but dysregulated activation can lead to harmful inflammation and tissue damage, which explains the effect of brensocatib.

The maximum recommended human dose (MRHD) for NCFBE is 25 mg brensocatib QD for chronic administration. At the MRHD, the steady state Cmax for total brensocatib based on population pharmacokinetic analyses is 0.62 µM (equivalent to 259 ng/mL) corresponding to an estimated human systemic free Cmax exposure of 0.079 µM.

In vitro mechanism of action studies using isolated recombinant human DPP1 enzyme showed that brensocatib competitively inhibited the activity of human DPP1 enzyme when studied in isolated form with an IC50 of 4.47 nm or using immortalized cell lines known to express DPP1 with an IC50 of 4.26 nm. (Study BS000449-19). The IC50 in the enzyme assay and the cell assay was similar, but the degree of inhibition caused by brensocatib is less in the U937 cell assay than observed in the isolated enzyme assay.

In an in vitro study to assess species selectivity, the inhibitory potency of brensocatib for purified recombinant DPP1 enzyme from human, rat and dog showed completely was measured. Brenscatib showed complete and concentration-dependent inhibition against recombinant DPP1 against all three species with IC50 against recombinant rat, dog and human enzyme of 21.4, 17.8, and 8.9 nM (pIC50 of 7.7, 7.8, and 8.05, respectively). (Study BS001015-29).

In another in vitro study to assess species selectivity, the inhibitory potency of brensocatib was measured for purified recombinant DPP1 enzyme from human, mice, rat and dog. In this study, brensocatib has similar IC50 against recombinant mouse, recombinant rat, dog, and recombinant human DPP1 enzyme of 24.1, 51.1, 38.1, and 17.1 nM, respectively. No direct effects on the enzymatic activities of NE, PR3, and CatG were observed (Study RES-0296).

It should be noted that the reported IC50 values are somewhat different between study RES-0296 and the studies BS000449-19 and BS001015-29. This finding is not critical but can be explained by the differences in the test conditions (substate concentrations, buffer pH, test temperature and test format, purity of the compound, etc.) and the performance in different laboratories and is described in study RES-0296.

Overall, the in vitro studies support the proposed mechanism of action, according to which brensocatib is an effective reversible inhibitor of recombinant dipeptidyl peptidase 1 (DPP1) in humans, mice, rats, and dogs, and confirmed the subsequent inhibition of NSP activities (NE, PR3, CatG) in human samples obtained from subjects treated with clinically relevant doses. The in-vitro data also support the selected test species for safety studies with regard to pharmacologically relevance

Additional MOA related information:

Importantly, there are data from DPP1 knockout (DPP1-/-) mice and phenotypes described in human subjects with a deficiency in DPP1, e.g. patients with Papillon-Lefèvre-Syndrom (PLS), a rare genetic condition with near-total to complete loss in DPP1 activity.

Studies in DPP1-/- mice have provided insights into the physiological role of DPP1 and the possible consequences of inhibiting the enzyme. DPP1-/- mice developed normally, appeared healthy, and were fertile. Histopathological examinations of the major organs, including the spleen, liver, lungs, heart, kidneys, thymus, brain, and bone marrow, revealed no obvious abnormalities, but are only reported as summary information (i.e., detailed information is not available) (Pham and Ley,1999, Adkison et al., 2002).

Human subjects with a deficiency in DPP1, typically present with two major clinical symptoms: severe palmoplantar keratosis, characterized by redness and thickening of the soles and palms, and periodontitis, which, if left untreated, is associated with tooth loss as early as the second or third decade in life (Biraggari et al, 2015; Toomes et al, 1999). There is also indication of enhanced bacterial skin and respiratory infections due to impaired neutrophil function and there might be mild immune system dysregulation, but there is no general dysfunction in the immune system.

Pharmacologically active metabolites

There is no specific information on pharmacologically active metabolites. In the Phase 1 mass balance and metabolic profiling study following a single 40 mg oral dose of [14C]-brensocatib to healthy male subjects, 6 metabolites were identified in plasma. M8 was the only major metabolite detected (51% of total AUC), and all other metabolites were minor (each < 0.5% of total AUC). Because M8 (thiocyanate) is an endogenous compound with known activities and all other metabolites were minor (each < 0.5% of total AUC), there is no need for further pharmacologically characterisation of human metabolites.

In Vivo

There is no in vivo animal model of bronchiectasis that provides direct evidence of the non-clinical efficacy of brensocatib. However, activation of neutrophils in the airways leads to the release of NSPs, including NE, which is thought to be central to the pathophysiology of bronchiectasis (Chalmers and Chotirmall, 2018). Therefore, the effect of brensocatib in the primary pharmacology studies was measured by the inhibition of DDP1 and the resulting reduction in NSP activity in neutrophil granulocytes from the bone marrow of in vivo treated animals as a surrogate marker for predicting efficacy in NCFBE.

The in vivo pharmacology of oral brensocatib administration has been evaluated in multiple mouse and rat strains covering a wide dose range.

Activation of NSP proenzymes by DPP1 occurs very early in the neutrophil maturation process (Palmer et al., 2018), when myeloblasts are transitioning to pro-myelocytes. Brensocatib would thus have no effect on NSP activity in the cells that have already progressed past the pro-myelocyte stage, including circulating neutrophils.

Initially the effect of DPP1 Inhibition by Brensocatib on NSP Reductions and PK Parameters was evaluated in single dose studies with brensocatib doses of up to 400 mg/kg in mice to understand the PK and PD profiles of brensocatib. Notably, in BALB/c mice, 24 hours after a single oral administration of up to 400 mg/kg brensocatib (vehicle control, 100, 200, and 400 mg/kg) no reductions in NPS activity in the bone marrow were observed, suggesting that neutrophil maturation and turnover take longer than one day. Surprisingly, in a similar experiment, brensocatib reduced NPS activity (NE, PR3, and CatG) in bone marrow lysates from male C57BL/6 mice after a single dose and at significantly lower exposure levels of up to 100 mg/kg (no vehicle control, 20, 60, and 100 mg/kg) in a dose-dependent manner.

Since DPP1 inhibitors are not expected to elicit an immediate reduction in systemic NSP activity, studies with daily oral administration of brensocatib for at least 7 days to mice and rats were conducted to allow

time for the effect on DPP1 in myeloblasts and subsequently time for complete turnover of circulating neutrophils. Incubation of bone marrow-derived neutrophil progenitor cells during their 7-day differentiation phase with brensocatib led to expected pharmacodynamic responses as represented by a concentration-dependent reduction in NSP activities in the cell lysates with inhibition potencies of 61.6, 208.9, and 114.8 nM for NE, PR3, and CatG, respectively. The extent of inhibition of DPP1 activity correlated with the reduction in NSP activities. The ability of the same lysates to degrade elastin, a key connective tissue component of the lung, was also inhibited. On average the greatest reduction was observed in CatG, while PR3 and NE were more moderately reduced relative to those mice receiving vehicle. In contrast, in various strains of rats, resulted in the greatest reduction in PR3, followed by CatG and then NE, relative to the vehicle control. The PK parameters of Cmax and AUC are reported from these studies. Overall, dose-dependent PK exposure responses (AUC and Cmax) were observed regardless of the rodent species and strain. Interestingly, DPP1 showed a gradual duration-dependent increase in activity when measured in the bone marrow.

The time to recovery of NSP activity/duration of pharmacologic effect on NSPs following termination of exposure to brensocatib was evaluated in two in vivo studies. Oral BID administration of brensocatib at 10 mg/kg/day in female mice was conducted for 8 days to achieve maximum NSP inhibition. Neutrophil serine protease activities were fully recovered compared to the vehicle control and a time to recovery of 6.6 days for NE, 7.5 days for PR3, and 5.8 days for CatG was calculated.

The in-vivo data also support the selected test species for safety studies with regard to pharmacologically relevance.

Transfer of the animal data to humans

Unfortunately, in terms of dose finding, optimisation and translation of the non-clinical data, there is some uncertainty of how much reduction in enzymatic activity of NSP enzymes (NE, CatG, and PR3) is required to achieve a relevant optimal effect, or how the preclinical PD results were translated into clinical dose finding in NCFBE. Although this is much more important in early clinical development and not necessarily critical after phase 3 as the preclinical pharmacology is overruled by the clinical data. Based on PK/PD modelling of brensocatib, the publication by Palmer (2018) states that 50% inhibition of NE in patients could be therapeutically relevant. The activity of NSP enzymes in plasma, blood and sputum was evaluated in clinical studies with a maximum reduction observed 7 to 10 days after the last dose with a reduction in NE activity of 39%, 54% and 65% at the 10, 25, and 40 mg dose levels, respectively, compared to 21% in the placebo.

However, the extent reduction of NSPs is also important for the functionality of neutrophils, which according to the clinical data should still have relevant NSP functionality in any case, even if the proportion of functional NSPs is reduced.

5.3.1.2. Secondary pharmacodynamics

The assessment of off-target effects of brensocatib (up to 10 µM) was investigated in the in vitro radioligand binding assay (168 potential target receptors). The most potent binding affinity was 1.6 µM against the 5-HT2A receptor and the most potent functional effect was 0.881 µM partial agonism of the Ghrelin receptor. The binding affinities to all targets (≥1.6 µM) are ≥94-fold lower than that for brensocatib towards human DPP1 (IC50 of 17.1 nM), indicating that brensocatib binding to DPP1 is highly selective. No adverse events were identified during the clinical trial programme that appeared to be related to an effect on the 5-HT2A receptor or the Ghrelin receptor (see clinical safety assessment).

In the in vitro electrophysiological assays to evaluate its potential for adverse cardiac effects, brensocatib showed only activity in channel, hCav3.2, with an IC50 value of 21.57 µM which is >1000-fold lower than that for brensocatib towards its DPP1 target.

Overall, based on an estimated human systemic free Cmax exposure of 0.079 µM at the maximum recommended human dose (MRHD) of 25 mg, the secondary pharmacodynamic data indicate little potential for off-target adverse effects at the systemic exposures likely to be achieved with brensocatib at the MRHD.

5.3.1.3. Safety pharmacology

The safety pharmacological effects of brensocatib on vital functions were adequately investigated in vivo core battery studies in cardiovascular (dogs), CNS (rats) and respiratory (rats) studies. In vitro studies comprised hERG inhibition assays. All pivotal safety pharmacology studies were conducted in compliance with GLP regulations and in accordance with ICH S7A and ICH S7B requirements (CPMP/ICH/539/00 and CPMP/ICH/423/02).

The cardiovascular safety study in Guinea Pigs has been performed in a GLP-compliant facility but does not claim GLP compliance. Since this study is not part of the core testing battery, it is not considered as pivotal, therefore it is considered acceptable.

It should be noted that only male animals (dogs, rats and guinea pigs) were tested in the in vivo safety studies. The core battery of safety pharmacological studies is particularly important in the early development phase of a drug and should generally be conducted prior to the first human exposure. Considering available preclinical and clinical data, which overall do not indicate any critical safety pharmacological effects, and as no critical sex-specific differences in pharmacokinetics/toxicokinetics were observed in dogs and rats, this issue will not be pursued further.

Brensocatib blocked the hERG-encoded potassium channel with an IC50 of 13.9 µM. The exposure margin to the free exposure (Cmax) at steady state of 0.079 µM (total Cmax = 0.616 µM or 259 ng/mL) in humans at MRHD (25 mg QD) is 176-fold, indicating a low arrhythmogenic risk. Brensocatib was further evaluated for secondary pharmaocologic effects in electrophysiological assays in a panel of 7 cardiac ion channels with no relevant in binding affinity at clinically relevant concentrations.

In the non-GLP-compliant cardiovascular study in anesthetized guinea pigs, no effects were observed at intravenous doses of up to 9.6 mg/kg. A safety margin has not been calculated by the applicant but can be estimated based on the total exposure (Cmax) at steady state in humans at MRHD of approximately 19-fold.

In the GLP-compliant telemetric study in the beagle dog there were no effects on cardiovascular parameters at 5 mg/kg but a small increase in the QA interval at 50 mg/kg (interval between the Q wave and the onset of the aortic blood pressure pulse as an indicator of cardiac contractility). Based on the results of this study, a NOEL (no observed effect level) of 50 mg/kg was established. However, even if one agrees that the small increase in the QA interval is not adverse, it is questionable to describe this increase as having 'no effect' at all. Therefore, the dose of 50 mg/kg was subsequently adjusted from NOEL (no observed adverse effect level) to NOAEL (no observed adverse effect level). The safety margin at the 50 mg/kg dose derived from the C_{max} in plasma is 43 (for free brensocatib) and 19 (for total brensocatib) relative to the MRHD. At 1000 mg/kg, there was an increase in the group mean HR accompanied by a transient decrease in the PR interval (the interval between the P wave (the onset of atrial depolarization) until the beginning of the QRS complex (the onset of ventricular depolarization)). An increase in the group mean diastolic and mean blood pressure (MBP), a decrease in the maximal rate of rise of the left ventricular pressure (dP/dtmax) and the left ventricular end diastolic pressure (LVEDP)

and an increase in the QA interval were also observed. At 1000 mg/kg, a small increase in the QT interval corrected for heart rate (up to 6%) was also observed, but there were no effects on QRS, waveform morphology, or body temperature.

Overall, the results of the telemetric study in the beagle dog are consistent with clinical findings where a single oral dose of brensocatib at 40 mg and 120 mg in healthy subjects showed negative effect on QTc prolongation and other cardiodynamic parameters (eg, HR, PR, QRS intervals, T-wave morphology, and U-wave presence).

In a GLP-compliant respiratory study in male rat, there were no apparent effects of brensocatib on respiratory parameters up to the highest tested dose of 200 mg/kg. Therefore, the NOEL for CNS effects is 200 mg/kg. Safety margin has not been calculated by the applicant but can be estimated based on the free exposure (Cmax) at steady state in humans at MRHD of approximately 72-fold.

In the GLP-compliant CNS study in male rat, there were no effects on neurobehavioural effects as measured by the Irwin test at brensocatib doses up to 200 mg/kg. Therefore, the NOEL for CNS effects is 200 mg/kg. Safety margin has not been calculated by the applicant but can be estimated based on the free exposure (C_{max}) at steady state in humans at MRHD of approximately 75-fold**.**

5.3.1.4. Pharmacodynamic drug interactions

No non-clinical pharmacodynamic drug interaction studies were conducted with brensocatib.

5.3.1. Pharmacokinetics

5.3.1.1. Absorption

The in vitro bidirectional permeability and efflux ratio of brensocatib was investigated in Caco-2 and MDCKII cellular monolayers.

In study ER20191540, brensocatib permeability in Caco-2 cell monolayers was evaluated as a part of a BCS classification assessment. Brensocatib was found to be highly soluble across a broad pH range. According to the Caco-2 permeability assay results, this drug was found to have high permeability; however, a significant amount of efflux (>3) was also noted, suggesting that active transporters are involved in removing this drug from the gut membrane (average Papp, a-b 7.17 × 10-6 cm/s and average Papp, b-a was 23.9 × 10-6 cm/s resulting an efflux ratio of 3.35).

In Study BJAA-0002-DV-PB, brensocatib permeability across Caco-2 monolayers was evaluated in the absence or presence of the efflux transporter P-glycoprotein (P-gp) inhibitors, verapamil (25 µM) or itraconazole (10 µM). Brensocatib (10 µM) was added to the donor chamber and incubated at 37°C for 120 minutes at pH 7.4. Digoxin (10 µM) with and without 25 µM verapamil and 10 µM itraconazole served as an efflux control, whereas 10 µM ranitidine (low permeability) and 10 µM warfarin (high permeability) served as permeability controls. The permeability coefficient Papp was determined by LC-MS/MS. The data from the controls were as expected. In the absence of the Pgp inhibitors, Papp, a-b was 9.03 × 10-6 cm/s with an efflux ratio of 3.86. In the presence of verapamil, Papp, a-b was 13.7 × 10-6 cm/s with an efflux ratio of 1.78 (efflux is inhibited 54.0%). In the presence of itraconazole, Papp, a-b was 12.9 × 10-6 cm/s with an efflux ratio of 2.38 (efflux is inhibited 38.4%). In conclusion, permeability data indicate that brensocatib is highly permeable and suggests that brensocatib is a Pgp substrate.

Overall, the permeability data indicate that brensocatib is highly permeable in MDCKII monolayers. The Pgp and BCRP efflux data suggest brensocatib is a Pgp and BCRP substrate.

Single-dose PK studies (non-GLP) were conducted in female mice (PO), male rats (IV and PO) and male dogs (IV and PO). In vivo brensocatib is highly bioavailable in male rats (75%) and male dogs (92%) following oral administration when compared to IV administration (single dose). No critical sex-specific differences in pharmacokinetics/toxicokinetics were observed in dogs and rats.

5.3.1.2. Distribution

Brensocatib binding to plasma proteins varied across species, with high binding in rats (93.9%), moderate binding in mice (83.5%) and human (87.2%), and low binding in rabbits (77.3%) and dogs (70.3%).

The in vitro binding of brensocatib to plasma protein from the male mouse, male rat, female rabbit, male dog and male human has been measured over a concentration range of 0.1 to 100 µmol/L brensocatib. In rat, an indication of a concentration dependent change in binding at concentration higher than 10 µmol/L was observed. The binding in rat was concentration dependent ranging from 6.47 at 10 µmol/L to 9.30 at 100 µmol/L. Between 0.1 and 10 µmol/L, although there was a trend between concentration of brensocatib and percentage unbound the fold change was small. The relevance for the assessment of the toxicology studies in rats and the safety margin calculation is regarded of low relevance considering the exposure data, particularly at the NOAEL and the percentage of change in unbound fraction.

There was negligible covalent binding potential of 14C-brensocatib binding following incubation in human hepatocytes (fraction covalently bound = 0.0188).

The average B/P ratios were 0.86 to 0.91 in male and female mice, 0.82 to 0.83 in male and female rats and 0.98 in dogs indicating low preferential binding of brensocatib to blood cells.

Tissue distribution in albino and pigmented rats using quantitative whole-body Autoradiography showed that Brensocatib was rapidly absorbed and widely distributed. The peak radioactivity tissue concentrations of most tissues were seen at 2 to 6 hours postdose with the highest concentrations in the liver and preputial gland in albino rats (radioactivity and tissue/blood ratio: 3.9 µg eq/g and 37.7 in liver at 2 hours; 3.54 µg eq/g and 39.6 in preputial gland at 6 hours) and in the uveal tract in the pigmented rats (radioactivity and tissue/blood ratio: 11.1 µg eq/g and 37 at 6 hours), suggesting melanin binding of brensocatib or its metabolites. At 672 hours (28 days) postdose in albino rats, radioactivity was below the limit of quantitation except for a few tissues, such as the liver, thyroid, kidney, spleen, adrenal gland. Given the short t½ of the parent drug in rats (~5 hours), the residual radioactivity is likely to be related to its metabolite(s).

Melanin-binding in the eye uveal tract and meningeal tissue:

The peak radioactivity tissue concentrations in the eye uveal tract and meningeal tissue of pigmented rats in the Quantitative Whole-body Autoradiography following oral administration of [14C]Brensocatib (2 mg/kg) were seen at 48 hours postdose with highest concentrations of 20.9 and 3.47 µg eq/g in the eye uveal tract and meningeal tissue (tissue/blood ratio: 1174 and 195). At the timepoint (168 hours postdose), concentrations had somewhat decreased, although remained at significant amounts with no clear elimination (concentrations of 17.5 and 2.09 µg eq/g in the eye uveal tract and meningeal tissue).

At the final timepoint (672 hours postdose) selected tissues from the rat head (Pigmented) following oral administration of [14C]Brensocatib (20 mg/kg) to male pigmented rats was 153.02 µg equiv/g and 22.85 µg equiv/g in the eye uveal tract and meningeal tissue. Terminal half-life of total radioactivity calculated for the uveal tract and meningeal melanin (using WinNonlin pharmacokinetic software) is 588 and 419 hours. Given the short t½ of brensocatib in rats (~5 hours), the residual radioactivity is likely to be related to its metabolite(s).

Excretion of brensocatib in the milk has not been specifically investigated in a distribution study. In the pre- and postnatal development study in rats brensocatib was detected in pups, suggesting that pups were likely exposed via maternal milk during lactation.

In consequence, based on the observed melanin binding in the fundus and meninges, a possible accumulation of brensocatib/brensocatib-related substances could therefore be expected. In both pigmented and albino rats the levels of radioactivity measured in the brain and spinal cord were low (≤0.032µg eq/g) and fell below the limit of quantitation (0.007 µg eq/g) at 48 hours postdose, indicating low CNS penetration.

Placental transfer of brensocatib-related radioactivity was shown in female time-mated albino rats. Peak radioactivity was observed 6 hours postdose in most maternal tissues. The peak radioactivity concentration in the amniotic sac and placenta were 3.762 µg eq/g and 0.448 µg eq/g. Tissue/blood ratios were 19.9 and 2.4. respectively. Peak levels in the whole fetus, fetus liver, and fetus eye were 0.14 µg eq/g (tissue/blood ratio: 0.7), 0.36 µg eq/g (tissue/blood ratio: 1.9) and 0.14 µg eq/g (tissue/blood ratio: 0.7), respectively.

5.3.1.3. Metabolism

The metabolism of brensocatib was evaluated in hepatocytes and intestinal microsomes from mice (intestinal microsomes only), rats, dogs, and humans. In hepatocytes, >95% remained as unchanged compound following a 4-hour incubation and a few minor metabolites were detected. The highest amounts of metabolites were detected in rat hepatocytes (1.9% for M1 and 0.9% for M4, both formed by oxidation). No unique human metabolite was identified.

In mouse, rat, dog, and human intestinal microsomes, 95.3%, 102%, 99.4%, and 100% remained as unchanged drug, respectively, after a 2-hour incubation.

Based on a cytochrome P450 (CYP) phenotyping experiment in E. Coli bacteria with over-expressed human CYP1A2, CYP2A6, CYP2B6, CYP2C8, CYP2C9, CYP2C19, CYP2D6, CYP2E1, CYP3A4, and CYP3A5. Among all CYP isozymes tested, only CYP2C8, 2D6, 3A4, and 3A5 were involved in brensocatib metabolism, responsible for 1.88%, 0.95%, 38.9%, and 58.3%, respectively, of brensocatib metabolism. Based on the results from this study, brensocatib was identified as a CYP3A4/5 substrate.

Metabolite profiling data in mice, rats, and dogs showed that brensocatib underwent metabolism via oxidation, N-demethylation, hydrolysis, N-formylation, oxidative dealkylation, hydration, dehydrogenation, and sulfuration. In mice, brensocatib was the most abundant component in plasma (71.9% of total AUClast). One major plasma metabolite, thiocyanate (M8), was detected (10.4% of total AUC) with Cmax of 0.249 µg eq/g, AUClast of 11.4 µg eq·h/g, and Tmax of 24 hours. All other circulating metabolites were minor < 10% of total AUC) or trace in abundance. In rats, brensocatib was the main component in plasma, urine (11.5% of dose), and feces (36.5% of dose), while M30 was the main component (6.4% of dose) in bile. In male and female plasma, brensocatib accounted for >80% of the total radioactivity exposure, and the most abundant metabolite (M8) thiocyanate was approximately 3% of total radioactivity exposure over 24 hours postdose (Cmax = 0.105 and 0.140 µg eq/g; AUClast = 0.849 and 1.04 µg eq*h/g in males and females, respectively). In dogs, the major metabolite in plasma was thiocyanate (M8), with Cmax and AUClast of 0.522 µg eq/g and 83.9 µg eq*h/g (67.1% of total AUC), respectively (plasma collected up to 240 hours). Brensocatib was the only other major component in plasma, with Cmax and AUClast at 1.33 µg eq/g and 29.2 µg eq*hr/g (23.4% of total AUClast), respectively, and was the only major component in urine (14.2% of dose). In feces, brensocatib and dioxybrensocatib (M17) were the major components at 25.7% and 14.8% of dose, respectively. All other metabolites were minor (<10%) to trace (<1%) in abundance.

Major human metabolite M8 (thiocyanate):

In a human mass-balance study, M8 (thiocyanate) was the only major metabolite (51% of total AUC in plasma but a trace urinary component (1.62% of the dose). All other metabolites were minor (each > 0.5% of total AUC).

The systemic exposure of thiocyanate (M8) in mice, rats and dogs at the doses evaluated in the mass balance study was either similar or higher than in humans following a single oral administration of 40 mg 14C-brensocatib in the mass balance study. Although thiocyanate was not directly measured in the repeated dose toxicity studies, it can be extrapolated from the doses used and the exposures measured in the mass balance studies that the NOAELs of 9 and 8 mg/kg/day in the chronic oral toxicity studies in rats and dogs cover the thiocyanate levels measured in the MHRD in humans.

Moreover, thiocyanate is an endogenous and a GRAS (generally recognized as safe) compound for the use as a component of the lactoperoxidase system, a common method for the preservation of raw milk/milk products). It's concentration in the blood stream can be influenced by smoking and consumption of dairy products and food with a normal range of 3 to 15 µg/mL.

Finally, clinical data showed that thiocyanate plasma concentrations were not affected by brensocatib dosing in healthy (≤120 mg single dose), renally impaired (25 mg single dose), and NCFBE (25 mg QD) subjects, i.e. corrected thiocyanate levels did not rise during QD administration of brensocatib 25 mg over 200 days.

Events of hypothyroidism, hypotension, and neurotoxicity were evaluated for their potential association with thiocyanate. No correlation was observed between thiocyanate concentrations and possible metabolite toxicity events.

5.3.1.4. Excretion

The biliary and urinary excretion of brensocatib were evaluated in intact and bile duct-cannulated (BDC) following oral or IV administration of 14C-brensocatib rats and intact dogs.

The orally dosed rats were divided into two groups of three males and three females. One of the groups had cannulae implanted in the bile ducts to collect urine, bile and faeces at 72-hour intervals, while the other group was left intact to allow urine and faeces samples to be collected over a longer period of 168 hours. The relative proportion of radioactivity from the original dose was determined in the faeces and cage washings of both groups by liquid scintillation counting. The mass-balance evaluation in intact and bile duct-cannulated rats revealed that following a single IV or oral dose of 14C-radiolabelled brensocatib to BDC rats, 15.5% to 23.7% and 19.0% of radioactivity were excreted mostly within 48 hours from urine and bile, respectively. In intact rats, the radioactivity recovery in feces was 76.8% following oral administration and 59.1% following IV administration.

The mass-balance evaluation in intact beagle dogs revealed that fecal excretion was the primary route of elimination of 14C-brensocatib-related radioactivity after an oral dose of 14C-brensocatib to male dogs, with a mean of 64.0% and of 18.0% of the dose recovered in feces and urine, respectively through 240 hours postdose.

The biliary and urinary excretion of brensocatib was also evaluated following IV administration of non-radiolabeled brensocatib in BDC rats and dogs. In BDC rats, 10.5% to 15.1% and 0.26% of dose was excreted in urine and bile as unchanged brensocatib, respectively. In BDC dogs after IV administration of non-radiolabeled of brensocatib, the renal clearance and biliary clearance were 19% and 3% of the total clearance, respectively.

Mass-balance studies in intact and bile duct-cannulated (BDC) rats and dogs with oral or IV administration of 14C-brensocatib indicate that brensocatib and its metabolites are mainly eliminated via fecal excretion while the parent drug is excreted moderately via urinary and minimally in bile.

5.3.1.5. Pharmacokinetic drug interactions

In vitro data indicate that brensocatib is a substrate of CYP3A, Pgp, and BCRP.

The IC50 for direct inhibition of CYP1A2, 2A6, 2B6, 2C8, 2C9, 2C19, 2D6, 3A4/5 and 2E1 and time-dependent inhibition of CYP1A2, CYP2C9, CYP2C19, CYP2D6 and CYP3A4/5 were above the maximum concentration of 30 µM and 50 µM respectively.

Brensocatib is a weak CYP3A inducer, but not an inducer of CYP1A2, CYP2B6, CYP2C8, CYP2C9, CYP2C19.

Brensocatib has little to no inhibitory effect on Pgp, BCRP, OATP1B1, OATP1B3, OAT1, OAT3, OCT2, MATE1, and MATE2K (IC50 >11 µM). Based on these data and plasma exposures at clinically relevant doses, brensocatib is unlikely to modulate the activity of CYP isozymes or drug transporters.

The results of the pharmacokinetic Drug-Drug Interactions (DDI) studies in relation to their clinical relevance with brensocatib was further evaluated in the clinical setting and will is discussed in the clinical assessment report. Both inducers and inhibitors of CYP3A and Pgp were studied in the Phase 1 program and further evaluated in PPK (population pharmacokinetic(s)) analysis and is discussed further in Module 2.7.2. Any potential clinical pharmacokinetic drug-drug interactions when using brensocatib will be addressed in the product's SmPC.

5.3.1.6. Other pharmacokinetic studies

Brensocatib has been fully characterized for pharmacokinetic properties in a program of in vitro and in vivo studies. No additional pharmacokinetic studies were warranted.

5.4. Toxicology

5.4.1. Single-dose toxicity

Stand-alone single-dose toxicity studies with brensocatib were not conducted. According to ICH guideline M3(R2) such information can be obtained from appropriately conducted dose-escalation studies or short-duration dose-ranging studies that define an MTD in the general toxicity test species. Brensocatib has been evaluated in non-GLP dose-escalation and short-duration dose-ranging studies that defined an MTD in the general toxicity test species. MTD (single administration) in rats and dogs was 1000 mg/kg. Since there is information on acute toxicity is available from other studies supported by appropriate GLP repeated-dose toxicity studies, the lack of standalone single-dose studies is acceptable.

5.4.2. Repeat-dose toxicity

Repeat-dose toxicity studies with oral administration of brensocatib were conducted for up to 6 months in rats and 9 months in dogs in compliance with GLP and ICH M3(R2) requirements.

Major findings/target organs are discussed and delineated below. Most of the findings were adequately reflected in section 5.3 of the proposed SmPC.

Mortality / early termination

DRF study in rats:

MTD phase: **premature death of 2 rats** (Day 3 repeated dosing of 1000 mg/kg/day) Exposure margins based on mean total/free AUC ~ 393/183.

RD phase: **premature death** (Day 7 following repeated dosing of 200 mg/kg) and **early termination** of all animals of this group (Day 9) due to adverse clinical signs. Exposure margins based on mean total/free AUC ~ 315/150.

6-Month toxicity study in rats: Two unscheduled deaths unrelated to administration of brensocatib (1M at 3 mg/kg/day (low dose) and one control M).

6-month study in dogs: severe periodontal disease (related to brensocatib) that necessitated the humane euthanasia of 4 out of 7 animals per sex in the high dose group (50 mg/kg/day).

Findings in the male reproductive tract (findings in the testis, epididymis and prostate)

Dose-dependent testicular toxicity was observed in dogs in the 1- and 6-month, but not 9-month, toxicity studies. In rat testicular toxicity was not observed. The male reproductive tissues in dogs in the dose-range finding study and in the 1- and 6-month GLP studies have been independently reviewed by an external expert.

In the dose range finding study, one dog dosed with 300 mg/kg/day for 14 days had an increase in epididymal cell debris but no changes in the testes. Based on the single animal group size and the frequent occurrence of cell debris in normal dog testes, this finding was not considered treatment related. In the 1-month study (animals were 8 to 9 months of age at dose initiation, at 0, 3, 15, or 75 mg/kg/day), testicular findings were noted at the high dose of 75 mg/kg/day only, which were broadly equivalent to those at 8 mg/kg/day in the 6-month study. Full reversibility of the testicular findings in the 1-month study was demonstrated after a 1-month recovery period.

In the 6-month study (5 to 6 months of age at dose initiation, at doses of 0, 2, 8, or 50 mg/kg/day), all dogs at 50 mg/kg/day showed marked to severe seminiferous tubular degeneration/atrophy with associated changes in the epididymides (decreased number of spermatozoa and/or increased cell debris and/or apoptosis on initial segment). The depleted seminiferous tubules were lined by Sertoli cells, which appeared to have healthy and viable basal nuclei. Leydig cell hyperplasia/hypertrophy was also seen at this dose, which was considered an adaptive response to decreased spermatogenesis. At 8 mg/kg/day, histopathological assessment of the testis showed similar though less severe changes, and there were no observations in Leydig cells. The pattern of changes in spermatogenesis suggests that the mechanism of toxicity targets spermatogonial and spermatocyte division and/or maturation as they progress through meiotic prophase. At 2 mg/kg/day (NOAEL), there were no brensocatib-related changes in the testes or epididymides. Following a 6-month dosing-free recovery period, substantial recovery in the high dose group was demonstrated by the presence of normal spermatogenesis in most of the seminiferous tubules and normal amounts of sperm in the epididymides, and the lack of any active degeneration of germ cells. However, recovery was considered incomplete due to variable degrees of hypospermatogenesis in these dogs (recovery was evaluated in control + high dose group only).

In order to characterize the potential toxicity of brenzocatib in dogs more precisely and, above all, to further investigate the dose-response relationship and the reversibility of the findings in the testicles and epididymis, a second chronic toxicity study (with 9 months of administration) was conducted in dogs with a lower maximum dose of 8 mg/kg/day and a recovery period of 26 weeks (similar duration to the previous study). In the 9-month study (7 to 8 months of age at dose initiation, at 0, 1, 2, 4, or 8 mg/kg/day), no testicular toxicity was observed at up to the high dose of 8 mg/kg/day (safety margins based on NOAEL and mean total/free AUC: 5/12). In addition, there were no changes in testosterone

levels or findings in full sperm evaluations. Overall, the exposure ranges are considered acceptable to ensure adequate clinical safety.

The age difference between the animals in the 6-month study and the 9-month study in dogs with regard to the results in the testes/epididymis and clinical safety in the youngest (pubertal) patients (aged ≥ 12 years) was a point of discussion. However, considering the totality of non-clinical and clinical data, this possibility is considered low overall and may be more relevant for dogs due to the high inter-individual variability in dogs.The findings noted in the testis from dogs (seminiferous tubule degeneration and atrophy) and epididymis (decreased number of spermatozoa and cellular debris) are mentioned adequately in the SmPC and the more conservative safety margin (5 times) was stated in section 5.3, which is considered acceptable.

Notably, no testicular finding was observed in any studies in rats or in the 28-day study in mice, despite much higher systemic exposures. Testicular toxicity was also assessed in the Phase 1 clinical study following administration of brensocatib at 40 mg QD for 28 days, and no clinically significant changes in spermatogram results were reported when evaluated at 13-week post last dose.

Periodontal disease:

In the 6-month study in dog study, administration of brensocatib at 50 mg/kg/day caused periodontal disease resulting in early termination of the group (safety margins based on the NOEL for periodontal disease (8 mg/kg) mean total/free AUC: 7/16). Periodontal disease is not an unexpected finding because human subjects with a deficiency in DPP1, suffer from severe periodontitis, which, if left untreated, is associated with tooth loss as early as the second or third decade in life (Biraggari et al, 2015; Toomes et al, 1999). No periodontal desease was observed at up to the high dose of 8 mg/kg/day in the 9-month study in dog study, (safety margins based on NOAEL and mean total/free AUC: 5/12).

Phospholipidosis

In the 1- and 6-month toxicity studies in rats and dogs, the morphological changes of vacuolated macrophages in the lung and to a lesser extent in the immune tissues including lymph nodes and spleen were noted. The vacuoles were believed to be a result of intracellular accumulation of phospholipids, which are typical for a condition described as PLD. The ultrastructural evaluation of the lung from brensocatib-treated animals with TEM demonstrated a classical pattern of whorls/lamellar lipid structures in these vacuoles, supporting the etiology of PLD. PLD is well known and described for a number of drugs and agents (Halliwell, 1997; Reasor, 1981; Reasor and Walker, 1981), typically being associated with the physicochemical property described as cationic amphiphilic (Chatman et al, 2009; Halliwell, 1997). Brensocatib contains a cationic hydrophilic cyclic amine moiety linked to a more lipophilic part, consistent with these properties. PLD is often prominent in the lung, related to a significant role of macrophages in phospholipid turnover in the lung (Halliwell, 1997). Vacuoles in other organs such as liver and kidney have also been commonly reported with agents causing PLD. The toxicological significance of PLD has been debated with its nature often described as adaptive rather than reflecting toxicity, and in most cases, it is reversible (Forbes et al., 2014; Lewis et al, 2014; Reasor, 1981; Reasor and Kacew, 2001; Reasor et al., 2006). In the toxicity studies with brensocatib, the PLD-related findings were dose-dependent and reversible. In rats, the changes were observed at ≥50 mg/kg/day in the lung and at 100 mg/kg/day in the lymphoid tissues, and no such effect was noted at up to 15 mg/kg/day. In the 1- and 6-month studies in dogs, the changes were observed at ≥8 mg/kg/day in the lung and at ≥50 mg/kg/day in the lymphoid tissues. In the 9-month dog study, no microscopic evidence of PLD was noted in any tissues at up to the highest dose of 8 mg/kg/day. The lung appears to be the most sensitive organ for brensocatib-induced PLD. Based on the changes in the lung, the corresponding exposure multiples relative to MRHD at the low and no observed effect levels for PLD are 150/71 (at 50 mg/kg/day in the 6-month study) and 28/13 (at 15 mg/kg/day in the 1-month study) in rats, and 7/16 and 5/12 at 8 mg/kg/day in the 6- and 9-month study in dogs, respectively. PLD is not considered monitorable

clinically. However, given the nature of the finding (vacuolated macrophages), a clear dose-response relationship, reversibility, and the safety margins, this microscopic finding is not expected to be clinically significant at the MRHD for NCFBE.

The argumentation presented above by the applicant can be accepted.

Lung (accumulations of vacuolated macrophages consistent with phospholipidosis)

In both rats and dogs, accumulation of vacuolated macrophages consistent with PLD was observed in the lung and was completely reversible. The low and no observed effect levels and corresponding exposure multiples for PLD in the lung are described in the Phospholipidosis section above.

Kidney changes

In the 28-day toxicity study in rats, an increased incidence of minimal kidney findings (tubular vacuolation) was observed in females at a dose of 15 mg/kg/day. As these kidney findings had no apparent effect on function, and partial recovery was observed at the 100 mg/kg dose (reversibility was investigated in the high dose group only), a NOAEL of 15 mg/kg was established. In the 6-month toxicity studies in rats and dogs, microscopic changes in the kidney associated with functional alterations as reflected in plasma and/or urinary parameters were identified in both species at 50 mg/kg/day (high dose). The same findings in the kidney were noted in the 1-month study in rats at 100 mg/kg/day and in dogs at 75 mg/kg/day. Safety margins based on NOAELs (kidney findings) and mean total/free AUC were 28/13 (1-month study in rats), 20/10 (6-month study in rats), 14/33 (1-month study in dogs) and 7/16 (6-month study in dogs). Therefore, the safety margin derived from the 6-month study in dogs represented the low-observed effect level for the kidney findings. Unfortunately, safety margins based on LOAEL and mean total/free AUC in the 6-month study in dogs was not calculable due to early termination of the 50 mg/kg/day dose group. Safety margins based on LOAELs (kidney findings) and mean total/free AUC were 199/94 (1-month study in rats), 150/71 (6-month study in rats) and 49/114 (1-month study in dogs). The changes in the kidney in the 1-month studies were partially reversible, and in the 6-month studies were fully reversible. Considering the reversibility results and the exposure multiples, the overall risk of renal toxicity in humans at MRHD for NCFBE is considered acceptable. Renal toxicity was not identified as a target for toxicity in the clinical programme.

Liver changes

In the 1-month study in dogs, reversible minimal to marked centrilobular vacuolation in the liver and increased levels of plasma ALT, AST, and GLDH were observed at 75 mg/kg/day (high dose). In the 6-month dog study, increased levels of ALT, AST, and GLDH were noted at 50 mg/kg/day, the highest dose evaluated, but without any microscopic correlates. Safety margins based on NOAEL and mean total/free AUC were 14/33 (1-month study in dogs) and based on NOAEL (liver changes) 7/16 (6-month study in dogs). In the 9-month study in dogs there were no changes in clinical chemistry parameters at dose levels up to 8 mg/kg/day (highest dose, safety margins mean total/free AUC were > 5/12). The applicant argues that other organs including the lung and lymphoid tissues were noted with a pattern of microscopic changes indicative of PLD, the hepatic responses may be related to this condition, as both vacuolated hepatocytes and increased plasma liver enzymes have been described in association with PLD (Asaoka et al., 2013; Hruban, 1984). However, a definitive correlation was not established. In the 1- and 6-month studies in rats, there were no noteworthy microscopic findings in the liver, although increases in ALT and AST values were noted in male rats at all brensocatib-treated doses and in a few individual females at the mid and high doses in the 6-month study.

Notably, elevated ALT and AST levels were still present in the 6-month toxicity study in rats at the end of the recovery period and in the 1-month toxicity study in rats, bile acids did not recover after a 4-week treatment-free period. As the elevated ALT and AST levels and the changes in bile acids were still present at the end of the respective treatment-free period, their classification as non-adverse was questioned.

The applicant clarified that the mean plasma ALT and AST levels in male animals showed a trend towards recovery, with only slight increases above the concurrent control values (1.5- to 2.1-fold) at the end of the treatment-free period, and that these sporadic changes in female animals were not considered test-item related, as minimal to mild increases in liver enzymes were observed in both control and brensocatib-treated female rats. With regard to changes in plasma bile acid observed after a four-week treatment-free recovery period, the applicant clarifies that the mean bile acid level in females at a dose of 100 mg/kg/day was slightly higher than in the concurrent controls (16.0 versus 6.2 µmol/L), but this was attributed to the lower values in the controls rather than to an actual finding related to the test object. Overall, the liver findings were classified non-adverse and of benign nature.

Immune System

Although some immune-related changes were observed in the nonclinical toxicity studies in rats and/or dogs (histological changes was the observation of PLD in the lymphoid tissues and periodontitis in dogs) they were reversible, dose-dependent, and occurred primarily at the highest dose evaluated in each study. In the completed clinical program, there have been no indications of brensocatib-related increases in the incidence of adverse events related to immunotoxicity or autoimmunity. The effect of brensocatib on neutrophil function and the microscopic changes consistent with PLD in the lymphoid tissues in rats and dogs are discussed as part of the assessment for potential immunotoxicity. Based on these comprehensive evaluations, brensocatib is deemed to have no potential for immunotoxicity at therapeutically relevant exposures.

Skeletal Muscle

Myofibre de-/re-generation in skeletal muscle was recorded in rats treated with 200 mg/kg/day for 7 days in the dose-range finding study, so a wider range of muscles was investigated in the 1-month GLP study. In the 1-month study in rats this effect was recorded at 100 mg/kg/day in soleus muscle only, which showed full reversibility. No histopathological signs of muscle damage were noted in the 6-month study in rats at up to the top dose of 50 mg/kg/day or in any studies in dogs. The corresponding exposure multiples relative to MRHD at the low and NOAEL for muscle toxicity in rats are 199/94 (at 100 mg/kg/day) and 150/71 (at 50 mg/kg/day), respectively.

It is agreed that given the exposure multiples, the finding in skeletal muscle in rats is not expected to be clinically relevant at the MRHD for NCFBE.

Findings related to the prostate:

In the 9-month toxicity studies in dogs, in one animal, minimal focal hyperplasia of the prostate was observed at a dose of 8 mg/kg/day (safety margins based on mean total/free AUC: 1/3). Since no signs of functional hyperplasia of the prostate were observed in any of the animals, this finding was not considered adverse. Focal hyperplasia of the prostate is probably part of the background pathology in dogs and occurs very rarely. Therefore, no clear relationship with the treatment could be established. Of note is that the group mean prostate weights in the main study were higher at ≥ 2 mg/kg/day. Since, there is considerable inter-animal variability in the weight of the prostate gland in dogs, this difference was was deemed to be unrelated to the test item. In the 6-month toxicity study in rats, a slight decrease in mean prostate gland weight was observed at 50 mg/kg/day. Given this change was not statistically significant, had no microscopic correlations, and reversed, it was considered non-adverse and does not support the prostate as a target organ of toxicity following brensocatib treatment in rats. Overall, the minimal and non-adverse effects of brensocatib in the prostate gland in nonclinical species in rats and dogs do not support the prostate gland as a target organ of toxicity.

5.4.3. Genotoxicity

A GLP-conform standard battery of *in vitro* and *in vivo* genotoxicity tests was performed with brensocatib in line with ICH S2(R1). Brensocatib was negative in an Ames test and in an *in vitro* mouse lymphoma TK assay (MLA) up to cytotoxic concentrations. Although there was a concentration trend for the increase in mutation frequencies (MF) +/- S9 mix at the 3-hour incubation time point and a slight statistically significant increase in MFs at one single concentration (80 µg/mL) in the MLA, this was considered not biologically relevant as it occurred at one concentration only and at high cytotoxicity levels (> 80%). There were no increases of micronuclei in bone marrow of rats in an *in vivo* micronucleus test up to 400 mg/kg given twice, which was considered the MTD based on a rat study with early mortalities at 1000 mg/kg/day after three days. In this study only 2000 immature polychromatic erythrocytes were analysed which is in line with the old version of the OECD 474 test guideline from 1997. As the study was performed in 2014 and hence before the new OECD 474 test guideline was established in 2016, where analysis of 4000 immature erythrocytes is recommended, this is not considered a concern. There was no bone marrow toxicity and brensocatib plasma levels were not analysed as evidence of exposure. However, there was a slight dose-related decrease in body weight. TK was bridged from a 28-day rat repeat-dose toxicity study. In this study the high dose of 100 mg/kg/day resulted in AUC_{free} of 116.35 µM*h which is 94-fold compared to the human AUC_{free} of 1.236 µM*h at MRHD (25 mg/day). Overall, the weight of evidence indicates that there is no genotoxic risk for patients.

5.4.4. Carcinogenicity

The carcinogenic potential of brensocatib was assessed in a 6-month Tg Ras mouse study and in a 104-week carcinogenicity study in rats. No neoplastic findings were observed in the Tg Ras mouse up to the highest dose of 50 mg/kg/day which resulted in AUC-based safety margins of 52-fold and 67-fold for total and free brensocatib at the MRHD of 25 mg/day, respectively.

In the 2-year rat study brensocatib treatment resulted in a marked and dose-dependent reduction in survival rates of females starting at 10 mg/kg/day. As a potential cause of death endometrial adenocarcinoma in the uterus/cervix (in 11 of 32 preterminal death) were identified in the 30 mg/kg/day group. However, the overall incidence of endometrial adenocarcinoma (preterminal death and terminal necroscopy) was not higher in brensocatib dosed groups as compared to control.

There was an increased incidence of endometrial adenoma in the high dose group (3/50; 6%) vs controls (1/50, 2%) also exceeding the historical range by this CRO in Han Wistar rats, which is maximum of 1 endometrial adenoma in 50 rats (2%). In addition, a dose dependent increase in the incidence of endometrial stromal polyps (1/50, 3/50, 5/50, 8/50 for control, 3, 10, 30 mg/kg/day, respectively) was observed.The trend test for endometrial stromal polyps was statistically significant but there was no statistically significant test item-related effect for the pair-wise comparison analyses in each dose group for stromal polyps. The lack of concern is further supported by the fact that the incidences of stromal polyps for all treated groups were within the range of the historical control data at the testing facility and there were no apparent dose-related trends for uterine stromal hypertrophy/hyperplasia or stromal sarcoma. When cervical and uterine tumours were combined (endometrial stroma, polyp/sarcoma and endometrial, adenoma/adenocarcinoma), no statistically significant trend was observed.

There was also a dose-dependent increased incidence of adenocarcinoma of the mammary gland (12% and 16% at 10 mg/kg/d and 30 mg/kg/d, respectively) above control (8%) and historical controls (range 2-10%) in female rats. However, there was no statistically significant trend (individual/combined) nor was statistical significance reached in pair-wise comparisons. It was further discussed that the incidences were only slightly higher than the historical control data, that there was absence of a dose-dependent pattern in the continuum of relevant proliferative or neoplastic changes and commonly reported

occurrences in Wistar Han rats. Furthermore, no similar neoplastic findings in the uterus, cervix, or mammary gland were reported in clinical studies with brensocatib.

Overall, the evaluation of malignancies in clinical studies revealed no pattern or dose-response relationship suggestive of a brensocatib-related effect on tumor development.

5.4.5. Developmental and reproductive toxicity

A complete evaluation of reproductive and developmental toxicity of brensocatib has been performed in line with the ICH S5 (R3) guideline.

Although no dedicated studies on fertility and early embryonic development (FEED) have been performed, these evaluations were included in the 6-months repeat-dose toxicity study in rats for male fertility and a definitive combined fertility and embryo-foetal developmental study in rats for female fertility.

With the exception of the dose range-finding study in non-pregnant rabbits, all studies were performed under GLP.

Toxicokinetic (TK) assessment was included in the reproductive and developmental toxicity studies.

Fertility and early embryonic development

Potential impact of brensocatib on male fertility was investigated in course of the rat 6-month repeat-dose toxicity study. During this study treated male animals were paired with untreated female animals. Administration of brensocatinib to male rats for 70 days at doses of 0, 3, 9 and 50 mg/kg/day had no effects on mating performance and fertility as indicated by the number of nights to a positive mating sign and by the number of animals pregnant. The number of pre- and post-implantation losses was similar across all groups.

Statistically significant, lower seminal vesicle gland weights (absolute and relative to terminal body weight) were observed in males dosed at ≥3 mg/kg/day and lower (absolute and relative to terminal body weight, not statistically significant) prostate weights were observed at 50 mg/kg/day, when compared with the control group. However, there were no correlating histological findings in these tissues or effects on male fertility, and these findings were not observed at the end of the recovery period. Therefore, the applicant did not consider these findings as adverse. The NOAEL for male fertility was set at 50 mg/kg/day which correspond to a systemic exposure in male rats of 97.0 µM and 1460 µM*h for C_{max} and $AUC_{0\text{-}24h}$, respectively. The safety margin to human exposure at the maximum recommended dose of 25 mg/day of brensocatib based on the AUC is 150-fold for total and 71-fold for free exposure.

In dogs testicular and epididymal degenerative changes were reported in the 6-month and 1-month studies at brensocatib doses of ≥8 mg/kg/day and 75 mg/kg/day, respectively with higher incidences and severity in prepubertal animals.

Potential impact of brensocatib on female fertility in rats was investigated in a combined fertility and embryo-foetal developmental study at dose levels of 0, 3, 20 and 100 mg/kg/day given orally 2 weeks prior to pairing and up until gestation day (GD)16. The treated female animals were paired with untreated male animals. There were no brensocatib-related effects on female estrous cycles, mating performance (including precoital interval), and fertility at any dose level. At the lowest dose of 3 mg/kg the number of pregnant animals was reduced due to male-associated impaired fertility. Also, the fertility index was lower in this group likely due to 3 females which were in oestrus on the first night of pairing and may have already ovulated. Therefore, both findings were not regarded as related to treatment with brensocatib. The NOAEL for effects on estrous cycles and female fertility was considered to be the top dose of 100 mg/kg/day which correspond to a systemic exposure of 84.4 µM and 1240 µM*h for C_{max}

and $AUC_{0\text{-}24h}$, respectively. The safety margin to human exposure at the maximum recommended dose of 25 mg/day of brensocatib based on the AUC is 128-fold for total and 61-fold for free exposure.

There were also no effects on female reproductive organs in repeated dose toxicity studies in rats and dogs.

Section 4.6/fertility of the SmPC currently states *"There are no fertility data in humans. Animal studies indicate no impact on male or female fertility (see section 5.3)."*

Embryo-foetal development

Potential effects of brensocatib on embryo-foetal development were investigated in rats and rabbits in dose-range findings and definitive EFD studies.

In rats this was done in a dose-range finding EFD study following exposure to brensocatib from GD6 to 16 as well as in a definitve combined fertility and EFD study following exposure to brensocatib from 14 days prior to pairing until up to GD16.

In the definitive fertility and embryo-foetal developmental study in rats administration of brensocatib resulted in slight maternal toxicity (e.g. changes in body weight gain and food consumption) at 20 or 100 mg/kg/day at the first days of treatment, only. Whereas no major malformations or external or visceral abnormalities/variations were observed, minor skeletal malformations of bent scapula and wavy ribs were noted after dosing with 100 mg/kg/day. There was an increased incidence of skeletal variations (malpositioned pelvic girdle and vestigial supernumerary full and/or short ribs in both cervical and thoracolumbar regions, incomplete ossification of sternebra) and differences in ossification after dosing with 20 or 100 mg/kg/day. After dosing with 3 mg/kg/day, there was reduced incidence of ossification in one skull bone only. An increased incidence of ossification of the phalanges (both forepaw and hindpaw) was seen at all dose levels of the hindlimb calcaneous at 20 or 100 mg/kg/day and of the interparietal skull at 100 mg/kg/day, indicating more advanced ossification in some fetuses in brensocatib-treated groups. The NOAEL for maternal toxicity and EFD was considered to be 3 mg/kg/day corresponding to a systemic exposure of 2.73 µM and 28.7 µM*h for C_{max} and $AUC_{0\text{-}24h}$ values on GD 16, respectively. The AUC-based safety margin to human exposure at the maximum recommended human dose of 25 mg/day is only 3-fold for total exposure and 1-fold for free exposure.

In **rabbits** potential effects of brensocatib on embryo/foetal development were investigated in a DRF-study and a definitive study.

In the DRF study doses of 15 and 50 mg/kg/day from GD7-GD19 had no effect on embryo/foetal survival or foetal development and induced only slight maternal toxic effects, like a transient decrease in food consumption and a decrease in body weight gain. In contrast, doses of 100 and 150 mg/kg/day were not tolerated by the F0 animals and resulted in premature termination of 3/6 and 4/6 animals, respectively, due to adverse maternal toxicity and evidence of abortion.

For the definitive EFD study in rabbits doses of 0, 5, 15 and 50 mg/kg/day were administered by oral gavage to rabbits from GD7 to 19. At doses of 15 or 50 mg/kg/day there was a slight maternal toxicity indicated by a slight reduction in body weight gain and some transient reductions in food consumption at 50 mg/kg/day only. There was no effect on embryo-foetal survival. In addition, there were no major malformations and no external or visceral abnormalities/variations. There was an apparent increase in several ossification parameters in the brensocatib-administered groups when compared to the control group (unossified/incompletely ossified odontoid process, cervical vertebral centrum/a and cervical vertebral arch(es) at 15 and 50 mg/kg/day along with cranial bone(s) small linear/linear ossification irregularity/ies and sacral vertebral centrum connected to sacro-iliac articulation centre at 50 mg/kg/day only). In addition, administration of brensocatib was associated with an increased incidence of pelvic girdle caudal displacement at 15 and 50 mg/kg/day and associated supernumerary ribs on the 13th

thoracic vertebra at all dose levels (significant at 15 and 50 mg/kg/day). Although supernumerary rib(s) occurred as the only observation within some fetuses, all fetuses with pelvic girdle caudal displacement (unilateral/bilateral) had at least one supernumerary rib, the longest, predominantly, being a "full" rib. According to the study report, as these variations occur commonly in this strain of rabbit, the observations recorded in this study are considered not to have any adverse consequence to the development of the rabbit fetus. Based on the results of this study, the NOAEL was considered to be 5 mg/kg/day for maternal effects and 50 mg/kg/day for embryo-foetal developmental effects. The corresponding mean C_{max} and $AUC_{0\text{-}24h}$ values at the embryofoetal NOAEL of 50 mg/kg/day on GD 19 of 23.0 µM and 195 µM*h, respectively, as determined in the preceding dose range finding study. The AUC-based safety margin to human exposure at the maximum recommended dose of 25 mg/day was 20-fold for total and 36-fold for free systemic exposure.

Prenatal- and postnatal development

The potential effect of brensocatib on pre- and postnatal development was investigated in rats after once daily oral dosing from GD6 to LD21 at dose levels of 0, 3, 9 and 20 mg/kg/day. As part of this study also the exposure of the weaning pups at PND/LD 4 was investigated. Brensocatib up to the highest dose level did not induce maternal toxic effects nor effects on pre- and postnatal development. Therefore, the NOAEL was set at the highest dose level of 20 mg/kg/day for F_0 maternal systemic toxicity, F_1 neonatal, developmental, systemic, and reproductive toxicity, and F_2 embryonic survival. At this dose level the systemic exposure was 14.1 µM and 163 µM*h for C_{max} and $AUC_{0\text{-}24h}$, respectively. The AUC-based safety margin to the maximum recommended human exposure at 25 mg/day is 17-fold for total and 8-fold for free exposures.

Studies in juvenile animals were not conducted. The 6-month repeated- dose toxicity studies were performed in adolescent rats and dogs (w age of the animals at the start of dosing: 6 to 7 weeks and 5 to 6 months, respectively). These studies are considered appropriate to support dosing of brensocatib in patients $\geqslant$12 years of age as the animals of both species entered puberty at the start of the studies (ICH, S11).

5.4.6. Toxicokinetics and exposure margins

The toxicokinetics (TK) of brensocatib were evaluated in mouse, rat, female rabbit, and dog.

Overall, in Han Wistar rats, brensocatib exposure increased in a dose-dependent manner with a small (< 2-fold). In dogs, exposure increased less than proportionally in the 14-day toxicity study (doses up to 300 mg/kg) but dose proportionally in the 1-month, 6-month, and 9-month toxicity studies (doses up to 75 mg/kg). No relevant sex differences in TK were observed. Accumulation was <2-fold following repeated dosing.

Steady state total/free AUC multiples relative to MRHD at the NOAEL are 28/13 (1-month toxicity study in rats), 20/10 (6-month toxicity study in rats), 14/33 (1-month toxicity study in dogs), 1/3 (6-months toxicity study in dogs) and 5/12 (9-month study in dogs).

Exposure margins to the NOAEL (male fertility) are 150/71 (6-month toxicity study in rats).

As the 9-month dog study represented a longer treatment duration and included more comprehensive assessment of testicular toxicity and male reproductive function, this study seems to be more relevant with respect to safety margins.

Overall, the exposure ranges obtained in the repeated dose toxicity studies are considered acceptable to ensure adequate clinical safety.

In the definitive EFD studies in rats and rabbits, plasma levels and TK parameters were not determined for all dose groups. The exposures from these dose groups were obtained from the DRF-EFD studies.

5.4.7. Local tolerance

Local tolerance was evaluated by histopathological assessments of the gastrointestinal tract in the repeat-dose toxicology studies in rats and dogs, following oral administration of brensocatib by gavage. There were no noteworthy histological findings in the gastrointestinal tract noted in any toxicology studies, although vomiting and fecal changes were noted in dogs with a dose-related increase in incidence and/or severity.

5.4.8. Other toxicity studies

Antigenicity

Brensocatib is a small molecule DPP1 inhibitor that has been fully characterized in a comprehensive program of toxicology studies. No antigenicity assessment was deemed necessary.

Immunotoxicity

Based on a weight of evidence approach in accordance with ICH S8 guideline, brensocatib did not reveal a concern for immunotoxic effects. Therefore, no additional nonclinical testing for immunotoxicity was considered necessary. A high-level summary of the assessment is provided as follows:

• The literature data indicate that DPP1-/- mice are normal, healthy, fertile, and maintain a preserved immune system and functionality. In addition, human subjects with PLS, a rare genetic condition with near total to complete reduction in DPP1 activity, typically present with 2 specific clinical symptoms consisting of palmoplantar keratosis and periodontitis; and they do not have general immune dysfunction. Therefore, inhibition of DPP1 with brensocatib is not expected to have a generalized adverse effect on the immune system.

Based on the pharmacological mechanism of action of brensocatib which is related and has a direct relevance to the regulation of the function of neutrophiles as part of the immunotoxicity:

While reduction of active NSPs may affect certain neutrophile function, studies in DPP1 knockout (DPP1-/-) mice and phenotypes described in subjects with PLS revealed no safety concerns or general dysfunction in the immune system. Studies conducted on DPP1-/- mice have provided insights into the physiological role of DPP1 and potential consequences resulting from inhibition of the enzyme. DPP1-/- mice developed normally, appeared healthy, and were fertile. Histopathological evaluations of major organs, including the spleen, liver, lung, heart, kidney, thymus, brain, and bone marrow, showed no apparent abnormalities. Flow cytometric analyses of various immune cell subsets, such as T cells, B cells, and natural killer cells derived from the spleen, thymus, lymph nodes, or blood, revealed no significant differences between DPP1-/- mice and wild-type mice. Although cytotoxic T lymphocytes derived from DPP1-/- mice showed defects in cell-mediated cytotoxicity in vitro, there was no evidence of infection with common mouse pathogens in the knockout animals (Pham et al,1999). Hematological analyses, including complete blood counts and differentials, indicated normal counts of circulating white blood cells and mature granulocytes in both wild-type and DPP1-/- mice. Cytospins of bone marrow cells derived from 4- to 8-week-old wild type and DPP1-/- mice revealed normal numbers of myeloid precursors and mature granulocytes. Although the absence of DPP1 severely reduced the activity of NE, CG, and PR3 in polymorphonuclear leukocytes, the lack of serine protease activity did not affect in vitro neutrophil chemotaxis or in vivo recruitment of leukocytes in response to thioglycollate, a nonspecific inflammatory stimulus (Adkison et al, 2002). In a collagen-induced arthritis mouse model, there were no differences

observed in humoral and cellular immunity between wild-type and DPP1-/- mice, as determined by measuring serum levels of IgG1 and IgG2a anticollagen antibodies and in vitro T-cell proliferation in response to collagen, respectively (Hu et al, 2005). Based on these nonclinical findings, it can be inferred that DPP1-/- mice maintained a preserved immune system and functionality.

In addition, human subjects with PLS, a rare genetic condition with near-total to complete loss in DPP1 activity, typically present with two clinical symptoms: severe palmoplantar keratosis, characterized by redness and thickening of the soles and palms, and periodontitis, which, if left untreated, is associated with tooth loss as early as the second or third decade in life (Biraggari et al, 2015; Toomes et al, 1999). Aside from these two specific phenotypic symptoms, there has been no report on general dysfunction in the immune system in those patients. Considering the pharmacological mode of action of brensocatib, the literature data that DPP1-/- mice maintained a preserved immune system and functionality, and the absence of general immune dysfunction in subjects with PLS, inhibition of DPP1 with brensocatib is unlikely to have a generalized adverse effect on the immune system.

• A database structure similarity search of PubChem, a comprehensive chemistry database containing over 115 million unique chemical structures and their associated bioactivities, and subsequent confirmatory visual inspections indicate that brensocatib does not resemble drugs that are known for immunomodulatory activity.

• In pivotal repeat dose general toxicity studies in rats and dogs, brensocatib did not cause any changes in globulins, albumin/globulin ratios, or the incidence of generalized infections. Dedicated assessment of neutrophil functions revealed a slight dose-related reduction in the ability of neutrophils to produce reactive oxygen species, but no effects on phagocytosis. Histologically, the immune-related changes consisted of the increased macrophage cellularity and/or accumulation of vacuolated macrophages in the lymphoid tissues, consistent with PLD. Overall, the immune-related changes in the nonclinical studies were dose-dependent, reversible, and occurred at higher doses with significant exposure multiples relative to the MRHD.

• In a rat quantitative whole-body autoradiography study, brensocatib-related radioactivity was not retained at high concentrations in the immune organs and tissues. The presence of trace radioactivity in these tissues was not considered toxicologically significant, given that in repeat dose general toxicity studies in rats and dogs the only immune-related histological changes was the observation of PLD in the lymphoid tissues which occurred at doses that provided significant multiples of the clinically relevant exposure.

• In the completed clinical studies, there have been no indications of brensocatib-related increases in the incidence of adverse events related to immunotoxicity or autoimmunity.

• The major metabolite of brensocatib, thiocyanate, is an endogenous compound and not associated with immunotoxicity, and treatment with brensocatib clinically had no impact on thiocyanate exposure.

Dependency

No dedicated nonclinical studies for assessing abuse/dependence were conducted, as a comprehensive nonclinical and clinical assessment did not reveal a concern. The assessment of abuse potential followed the US FDA Guidance for Industry on the Assessment of Abuse Potential of Drugs (FDA, 2017) and the EMA Guideline on the Nonclinical Investigation of the Dependence Potential of Medicinal Products (EMA, 2006). A high-level summary of the assessment is provided as follows:

Based on pharmacological mechanism of action (literature data in DPP1-/- animals and phenotypes described in humans with PLS), structural similarity database searches and confirmatory visual inspections (brensocatib does not resemble the structures of common drugs), IC50 values for potential targets determined in an in vitro CEREP off-target screening assay, negligible CNS exposure, results of

the special CNS safety pharmacology study and the general repeated-dose toxicity studies in rats and dogs (in which no clinical signs suggestive of potential CNS-specific effects on abuse or dependence were observed) and results reported in the clinical studies with brensocatib (no reported events falling within the narrow terms of the standardised MedDRA query for drug abuse, dependence and withdrawal). The major metabolite, thiocyanate, is an endogenous compound not associated with abuse potential.

Studies on metabolites

No specific non-clinical studies in order to qualify metabolites were conducted.

M8 (thiocyanate) is the only major human metabolite identified in human subjects following administration of brensocatib at a 40 mg dose (maximum possible clinical dose for all indications under evaluation). Thiocyanate is also an endogenous compound in humans and it is present in nonclinical toxicology species conducted with brensocatib. In a clinical study thiocyanate plasma concentrations were not affected by brensocatib dosing in healthy (≤120 mg single dose), renally impaired (25 mg single dose), and NCFBE (25 mg QD) subjects, i.e. corrected thiocyanate levels did not rise during QD administration of brensocatib 25 mg over 200 days.

Studies on impurities

The nitroso impurity was identified in the brensocatib drug substance and tablet drug product. In order to test mutagenicity, an GLP-conform enhanced Ames test (EAT) was performed which yielded a negative result. To follow up on the negative EAT result, a mammalian cell mutation assay using 5% rat S9 mix was performed. This assay was clearly negative. Unfortunately, it was not performed with hamster S9 mix, at least at low concentrations as it is known to be cytotoxic for many cell cultures. In addition, no nitrosamine positive controls were included in the study. Finally, metabolism data using human microsomes and rat S9 mix indicated a low potential of α-hydroxylation of the nitroso impurity, however the α-hydroxylation pathway and potential formation of a diazonium ion could not completely be ruled out. An evaluation using quantum mechanics (QM) Computer-Aided Discovery and Redesign (CADRE) model indicates that the nitroso impurity has a low carcinogenicity potency. , with a predicted TD_{50} >1.5 mg/kg. Although the CADRE model predicts that several P450 isozymes (CYP1A1, 2A6, 2B6, and 2C8) have the potential for catalytically optimal binding and α-hydroxylation of the nitroso impurity, the CADRE potency prediction takes precedence, in the framework of the CADRE QM carcinogenicity potency and P450 binding evaluations. These data indicate that the nitroso impurity has a low carcinogenic potential, which further justifies a limit of 1.5 µg/day.

Taken together, the whole body of evidence suggests, if there is any mutagenic potential, it is probably low. During the procedure CHMP has received a response from NSOEG, which also concluded that nitroso impurity can be controlled at 1.5 µg/day.

Other mutagenic impurities are adequately controlled in accordance with ICH M7(R2). The non-mutagenic impurities are considered toxicologically qualified above the qualification threshold in accordance with ICH Q3 guidance.

In the 9-months toxicity study, an impurity was also tested at 0.06 mg/kg/day in all brensocatib-treated groups from Weeks 36 to 39. Therefore, this impurity was considered qualified at the level of 0.06 mg/kg/day.

Phototoxicity studies

Brensocatib absorbs light in the wavelength range of 290 to 700 nm, but without a peak above 290 nm. Furthermore, a whole-body autoradiography investigation of disposition in pigmented and non-pigmented rats showed significant distribution to uvea and other melanin containing tissues in pigmented rats. Consequently, the potential for phototoxicity of brensocatib was further investigated. Brensocatib was evaluated for phototoxic potential using an in vitro neutral red phototoxicity (3T3 cell) assay. The

results of this assay showed that brensocatib was not phototoxic in this in vitro test system when tested up to the limit of solubility and stability in the primary vehicle, according to the OECD guideline.

Excipients studies

There are no novel excipients.

Mechanistic studies

Histopathological evaluation of tissues from the 1-month GLP studies in rats (Study No. 527409) and dogs by light microscopy demonstrated vacuoles in different organs suggestive of possible intracellular phospholipid accumulation. To further elucidate the characteristics of the observed vacuoles in lungs and other organs, lungs from selected animals (negative controls in the vehicle groups and animals with distinct vacuoles in the high dose groups) in the 1-month repeat-dose toxicology studies in rats and dogs, as well as kidney from rats, ultrastructural evaluation of lung and kidney tissues were investigated with transmission electron microscopy (TEM). Samples evaluated included slices of formalin-fixed lung from 2 control rats and 2 rats dosed at 100 mg/kg/day and kidney from 4 control rats and 4 rats dosed at 100 mg/kg/day, and lung from 2 control dogs and 2 dogs dosed at 75 mg/kg/day.

The ultrastructural evaluation of photomicrographs of lung from brensocatib-treated rats revealed an increase in size of alveolar macrophages and increased lysosomal accumulation of definitive whorls of membrane remnant material. This was considered to be consistent with PLD. Similarly, increased lysosomal accumulation of membrane remnant material of alveolar macrophages in dog lung was consistent with PLD. Ultrastructural evaluation of photomicrographs of kidney from brensocatib-treated rats revealed predominantly low-grade degenerative changes of mitochondrial swelling, increased luminal debris, and in females only cytoplasmic vacuolation or cytosolic rarefaction. In the kidney of brensocatib-treated males, no definitive ultrastructural finding in examined regions was observed to correlate with the light microscopy findings of tubular vacuolation; however, in females, potential ultrastructural correlates for renal tubular vacuolation seen on light microscopy examination included proximal tubule cytoplasmic vacuolation and/or proximal tubule cytosolic rarefaction.

5.4.9. Ecotoxicity/environmental risk assessment

The applicant provided a detailed Phase I & Phase II assessment for the active ingredient brensocatib.

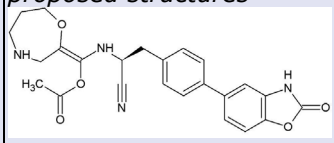
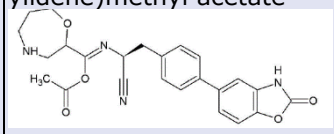
Table 3: Summary of main study results: Phase I

Substance (INN/Invented Name):	*brensocatib*		
CAS-number (if available):	*1802148-05-5*		
PBT/vPvB screening			
Study type	Test protocol	Result	Conclusion
Bioaccumulation potential-log Kow	*OECD107*	*-0.753 at pH 5* *0.837 at pH 7* *1.77* at pH *9*	Potential PBT: *N*
PBT/vPvB assessment			
Property	Parameter	Result	Conclusion
Bioaccumulation	*log Kow*	*0.837* at pH 7	*Potentially not B*
Persistence	Ready biodegradability	*N*	*potentially P*
	$DT_{50,Total\ System}$ at 12°C	*⩾10000* d	*vP*
Toxicity	$NOEC_{aquatic\ (Daphnia)}$ C, M, R STOT RE 1 or 2*	*0.37* mg/L	*not T* *potentially T*
PBT/vPvB statement:	Brensocatib is considered to be not PBT, nor vPvB		

Phase I			
Parameter	Value	Unit	Conclusion
PEC_{sw}, *default*	0.125	µg/L	≥ 0.01 threshold: *Y*
Other concerns (e.g. chemical class)			*N*

Table 4: Summary of main study results: Phase II

Phase II Physical-chemical properties and fate			
Study type	Test protocol	Result	Remarks
Water solubility	OECD 105	*945* mg/L at *20 °C (pH 5)* *71.1* mg/L at *20 °C (pH 7)* *10.1* mg/L at *20 °C (pH 9)*	*shake flask*
Dissociation in Water	OECD 112	$pK_{a,\ 1}$ = $pK_{a,\ 2}$ =	Not conducted
Adsorption-Desorption Soil 1 = *loam*	OECD 106	 $K_{FOC,\ soil\ 1}$ = *66900* L/kg_{oc}	
Soil 2 = *loam*		$K_{FOC,\ soil\ 2}$ = *27600* L/kg_{oc}	
Soil 3 = *silt loam*		$K_{FOC,\ soil\ 3}$ = *35200* L/kg_{oc}	
Sludge 1 = *municipal*		$K_{FOC,\ sludge\ 1}$ = *344* L/kg_{oc}	
Sludge 2 = *municipal*		$K_{FOC,\ sludge\ 2}$ = *984* L/kg_{oc}	
Ready Biodegradability Test	OECD 301B	0 % (28 d) not readily biodegradable	
Aerobic and Anaerobic Transformation in Aquatic Sediment systems Sediment 1 = sandy loam	OECD 308	$DT_{50,\ water\ 1}$ = 0.8d $DT_{50,\ sediment\ 1}$ ≥10000 d $DT_{50,\ whole\ system\ 1}$ = ≥10000 d CO_2 = 16 % NER_{max} = 43.8 % $NER_{testend}$= 40.5%	20°C
Sediment 2 = sand		$DT_{50,\ water\ 2}$ = 1.1 d $DT_{50,\ sediment\ 2}$ = ≥10000 d $DT_{50,\ whole\ system\ 2}$ = 198 d CO_2 = 13.2 %	20°C

		NER_{max} = 58.2 % $NER_{testend}$= 58.2%	
Transformation products		>10% = Y TP1 (max) = 33.5 % at day 2, total system	*proposed structures*  (E)-{[(1S)-1-cyano-2-([2-oxo-1,3-benzoxazol-5-yl]phenyl) ethyl]amino}(1,4-oxazepan-2-ylidene)methyl acetate  (Z)-{[(1S)-1-cyano-2-([2-oxo-1,3-benzoxazol-5-yl]phenyl) ethyl]imino}(1,4-oxazepan-2-yl)methyl acetate

Phase II Aquatic effect studies					
Study type	**Test protocol**	**Endpoint**	**Value**	**Unit**	**Remarks**
Algae, Growth Inhibition Test/*Raphidocelis subcapitata*	OECD 201	NOEC	2300	µg/L	growth rate
Daphnia sp. Reproduction Test/ *Daphnia magna*	OECD 211	NOEC	370	µg/L	growth (body length)
Fish, *ELS/Pimephales promelas*	OECD 210	NOEC	940	µg/L	hatching success, survival, growth
Activated Sludge, Respiration Inhibition Test	OECD 209	NOEC	≥1x10^6	µg/L	total respiration
Phase II Sediment effect studies					
Sediment Dwelling Organism Test/*Chironomus riparius*	OECD 218	NOEC	150	mg/kg_{dw}	combined development rate, 1.9% o.c., no correlation

Risk characterisation				
Compartment	**PEC**	**PNEC**	**RQ**	**Conclusion**
STP	1.25 µg/L	1x10^5 µg/L	1.25x10^{-5}	No risk
Surface water	0.125 µg/L	37 µg/L	0.003	No risk
Groundwater	0.031 µg/L	3.7 µg/L	0.0084	No risk
Sediment	0.273 mg/kg_{dw}	1.5 mg/kg_{dw}	0.182	No risk

A bioaccumulation potential is not indicated based on the log K_{OW} < 4.5. A definitive PBT/vPvB assessment is not required. Based on the results of the water-sediment study (OECD 308) brensocatib has to be classified as very persistent in the environment.

Considering the above data from Phase I and Phase II, brensocatib is not expected to pose a risk to the environment.

5.5. Overall discussion and conclusions on non-clinical aspects

5.5.1. Discussion

Pharmacology

In vitro studies support the proposed mechanism of action, according to which brensocatib is an effective reversible inhibitor of recombinant dipeptidyl peptidase 1 (DPP1) in humans, mice, rats, and dogs, and confirmed the subsequent inhibition of NSP activities (NE, PR3, CatG) at clinically relevant doses.

Primary pharmacology

Brensocatib is a selective, reversible inhibitor of didpetidyl peptidase I (DPP1) at low nanomolar concentrations. Brensocatib has similar IC50 against recombinant mouse, recombinant rat, dog, and recombinant human DPP1 enzyme of 24.1, 51.1, 38.1, and 17.1 nM, respectively. DPP1 is an enzyme that is responsible for the activation of NSPs in the bone marrow during the maturation of neutrophils. *In vitro* data also confirmed the subsequent inhibition of NSP activities (NE, PR3, CatG) and no direct effects of brensocatib on the enzymatic activities of NE, PR3, and CatG were observed.

There is no need for further pharmacological characterisation of M8 (thiocyanate), the only major human metabolite. Thiocyanate is an endogenous compound with known activities.

There is no *in vivo* animal model of bronchiectasis that provides direct evidence of the non-clinical efficacy of brensocatib. However, activation of neutrophils in the airways leads to the release of NSPs, including NE, which is thought to be central to the pathophysiology of bronchiectasis. Therefore, the effect of brensocatib in the primary pharmacology studies was measured by the inhibition of DPP1 and the resulting reduction in NSP activity in neutrophil granulocytes from the bone marrow of *in vivo* treated animals as a surrogate marker for predicting efficacy in NCFBE, i.e. as a surrogate for the lack of an experimental animal model. *In vivo* pharmacology studies with daily oral administration of brensocatib to mice and rats have shown decreased NSP activities in bone marrow lysates but also increased DPP1 activity. Since an increased activity of DPP1 had no impact on the ability of brensocatib to reduce NSP activity across rodent studies, this finding is not pursued further.

The time to full recovery of NSP activity after cessation of dosing is 6-8 days in female mice.

It is noteworthy that the nonclinical *in vitro* and *in vivo* finding overall translates to clinical findings where QD administration of brensocatib at 10 to 40 mg (steady state free Cmax exposure of 0.079 µM (equivalent to 259 ng/mL) at MHRD of 25 mg) led to a significant reduction in the activity of NE, PR3, and CatG in blood among healthy, NCFBE and cystic fibrosis (CF) subjects, and in sputum samples from NCFBE and CF subjects.

The pharmacology data also support the selected test species for safety studies with regard to pharmacologically relevance.

Secondary pharmacology

Off-target effects of brensocatib (up to 10 µM) were investigated in a battery of 168 potential target receptors and *in vitro* electrophysiological assays to evaluate potential adverse cardiac effects. Overall, selectivity of brensocatib has been shown and the data indicate little potential for off-target adverse effects at the systemic exposures likely to be achieved with brensocatib at the maximum recommended human dose (MRHD).

Safety pharmacology

In vitro hERG-encoded potassium channel assay is indicating a low arrhythmogenic risk (176-fold safety margin). Pivotal CNS and respiratory safety studies in rats suggest a low risk for adverse effects (safety margins > 72 fold). Overall, the *in vivo* safety pharmacology studies indicate that brensocatib may cause some cardiovascular effects. In the GLP-compliant telemetric study in the beagle dog there were no effects on cardiovascular parameters at 5 mg/kg but a small increase in the QA interval at 50 mg/kg. Based on the results observed in the non-clinical studies in male dogs, and the safety margins between NOAEL (50 mg/kg) and C_{max} at the MRHD (19-fold for total and 43 for free brensocatib) there are actually

only limited concerns regarding cardiovascular effects. It should however be noted that in the clinical trials program, where a single oral dose of 40 mg and 120 mg brensocatib was administered to healthy subjects negative effect on QTc prolongation and other cardiodynamic parameters were observed (eg, HR, PR, QRS intervals, T-wave morphology, and U-wave presence), therefore the PI does not need to reflect the non clinical findings.

Pharmacodynamic drug interaction

No non-clinical pharmacodynamic drug interaction studies were conducted with brenzocatib. There are currently no approved drugs which compete with brensocatib at the pharmacological target (DPP1 and downstream NSPs) and/or have similar or opposing pharmacodynamic effects.

Pharmacokinetics

Appropriately validated LC-MS/MS analytical assays for the quantification of brensocatib were used to support non-GLP and GLP studies.

The absorption properties of brensocatib were evaluated *in vitro* in Caco-2 and MDCKII cells and *in vivo* in mouse, rat and dog PK studies following single IV or oral administration or in toxicology studies following repeated once-daily oral administration. Brensocatib is highly permeable *in vitro* and highly bioavailable in rats (75%) and dogs (92%) following oral administration when compared to IV administration (single dose) following oral administration in rats and dogs.

The Pgp and BCRP efflux data suggest brensocatib is a Pgp and BCRP substrate.

Brensocatib binding to plasma proteins varied across species, with high binding in rats (93.9%), moderate binding in mice (83.5%) and human (87.2%), and low binding in rabbits (77.3%) and dogs (70.3%). 14C-brensocatib has negligible covalent binding potential in human hepatocytes and the B/P indicates low preferential binding to blood cells.

Tissue distribution in albino and pigmented rats using quantitative whole-body Autoradiography showed that brensocatib was rapidly absorbed and widely distributed. Highest radioactivity tissue concentrations were in the liver and preputial gland in albino rats and in the uveal tract in the pigmented rats, suggesting melanin binding of brensocatib or its metabolites. There is low CNS penetration. Placental transfer of brensocatib-related radioactivity was shown in female time-mated albino rats. Excretion of brensocatib in the milk has not been specifically investigated in a distribution study, however, in the pre- and postnatal development study in rats brensocatib was detected in pups, suggesting excretion in maternal milk. Significant melanin binding in the eye uveal tract and meningeal tissue with remaining at significant amounts with no clear elimination at 168 hours and even 672 hours postdose. Based on data from observations and exposure margins in chronic toxicity studies (at least 5 times (dogs) and 150 times (rat) the MRHD of 25 mg/day on an AUC basis), the potential toxicity risk associated with the accumulation of brensocatib or brensocatib-related material in the uveal tract and meningeal or surrounding tissues is considered minimal. There is no comparable binding to pigmented skin structures.

In vitro, slow metabolism of brensocatib was detected in rat, dog, and human hepatocytes (96.4%, 99.7% and 100% remaining). The few minor metabolites that were detected (each less than 2% of total radioactivity) were mediated by cytochrome P450 (CYP)3A4/5 and no unique human metabolite was identified.

In a human mass-balance study, M8 (thiocyanate) was the only major metabolite. The available data from mice, rats and dogs show that animals were exposed to similar or higher circulating M8 concentrations than humans following brensocatib administration. Thiocyanate is an endogenous compound with a significant background plasma spectrum, and clinical data have shown that thiocyanate plasma concentrations of human subjects treated in clinical trials were not affected by the administration of brensocatib in the form of QD for 200 days. Therefore, CHMP considered that the human 'major'

metabolite thiocyanate (M8) can be considered qualified and no further non-clinical studies are required for qualification. However, the proposed metabolism of brensocatib was subject to some uncertainty because the the radiolabel was seemingly located in an unstable position which, when removed, forms thiocyanate, a small molecule which is not representative of brensocatib metabolism. It seemed that a larger portion of brensocatib molecule remains metabolized in unknown ways after the radiolabelled smaller part is removed. This is especially applicable in dogs, where the M8 metabolite represents 67.1 % of total AUC. The applicant provided a discussion on metabolism characterization of brensocatib and why it is believed that the radiolabel was put in a stable position. Although M8 formation leads to loss of the ^{14}C label, it occurs slowly, compared to brensocatib. While M8 represented a large proportion of plasma AUC due to its long half-life, it accounted for <1% of the excreted dose, suggesting limited overall formation. Furthermore, any metabolites formed after nitrile cleavage would be inactive since nitrile moiety is necessary for binding to DPP1. In conclusion, the human mass balance study is considered sufficient for brensocatib metabolism characterization.

The biliary and urinary excretion of brensocatib were evaluated in rats and dogs. Brensocatib is moderately excreted in urine and bile and mostly excreted in feces. The PK DDI potential of Brensocatib was evaluated *in vitro*. The IC50 for direct inhibition of CYP1A2, 2A6, 2B6, 2C8, 2C9, 2C19, 2D6, 3A4/5 and 2E1 and time-dependent inhibition of CYP1A2, CYP2C9, CYP2C19, CYP2D6 and CYP3A4/5 were above the maximum concentration of 30 µM and 50 µM.

In conclusion, i*n vitro* studies are inconclusive regarding the potential of brensocatib to induce CYP2B6 and CYP3A4 (see section 5.2). The SmPC adequately reflects the conclusion reached as follows:

In vivo induction cannot be excluded. Co-administration with CYP3A4 substrates used in bronchiectasis (e.g. inhaled corticosteroids, macrolide antibiotics or inhaled bronchodilators such as salmeterol or vilanterol) may result in decreased plasma concentrations and reduced therapeutic effect. Adjustment of the concomitant treatment may be considered if efficacy is reduced.

Brensocatib is not an inducer of CYP1A2, CYP2C8, CYP2C9, CYP2C19. Brensocatib has little to no inhibitory effect on Pgp, BCRP, OATP1B1, OATP1B3, OAT1, OAT3, OCT2, MATE1, and MATE2K (IC50 >11 µM). Based on these data and plasma exposures at clinically relevant doses, brensocatib is unlikely to modulate the activity of CYP isozymes or drug transporters.

Toxicology

Single-dose toxicity

The use of rats (Han Wistar) and dogs as animal models for toxicological studies was adequately justified.

Single dose toxicity studies with brensocatib have not been conducted, which is justified. Single dose toxicity was addressed in MTD/dose finding studies conducted in rats and dogs. The MTD (single dose) in rats and dogs was 1000 mg/kg. In the rat study, mortality was observed, and some rats were killed prematurely for welfare reasons, but at doses far more than any clinical relevance.

Repeat-dose toxicity

Repeat-dose toxicity studies with oral administration of brensocatib were conducted for up to 6 months in rats and 9 months in dogs in compliance with GLP and ICH M3(R2) requirements. The pivotal studies were preceded by MTD / DRF studies in rats and dogs. Overall, an adequate safety profile with sufficient safety margins was established. Major targets organs of toxicity were identified in rats and dogs. In the 6-month studies, microscopic changes were noted in the lung and kidney in both species, and additionally in lymphoid tissues, testes/epididymides, and periodontal tissues in dogs. No noteworthy findings were noted in the 9-month study in dogs. All brensocatib-related findings were usually dose-dependent, reversible (dependent on dose and recovery time), and occurred at exposures greater than at the exposures at the MRHD. As the 9-month dog study represented a longer treatment duration and included

more comprehensive assessment of testicular toxicity and male reproductive function, this study seems to be more relevant with respect to safety margins. Notably, no testicular finding was observed in any studies in rats or in the 28-day study in mice, despite much higher systemic exposures and no clinically significant changes in spermatogram results were reported in a Phase 1 clinical study. In a preceding 6-month dog study, administration of brensocatib at 50 mg/kg/day caused periodontal disease resulting in early termination of the group. Also notable is that periodontal disease is not an unexpected finding because human subjects with a deficiency in DPP1, suffer also from severe periodontitis.

In the 28-day toxicity study in rats, an increased incidence of minimal kidney findings (tubular vacuolation) was observed in females at a dose of 15 mg/kg/day. As these kidney findings had no apparent effect on function, and partial recovery was observed at the 100 mg/kg dose (reversibility was investigated in the high dose group only), a NOAEL of 15 mg/kg was established.

Elevated ALT and AST levels were still present in the 6-month toxicity study in rats at the end of the recovery period and in the 1-month toxicity study in rats, bile acids did not recover after a 4-week treatment-free period. As the elevated ALT and AST levels and the changes in bile acids were still present at the end of the respective treatment-free period, their classification as non-adverse was questioned. The applicant clarified that the mean plasma ALT and AST levels in male animals showed a trend towards recovery, with only slight increases above the concurrent control values at the end of the treatment-free period, and that these sporadic changes in female animals were not considered test-item related, as minimal to mild increases in liver enzymes were observed in both control and brensocatib-treated female rats. With regard to changes in plasma bile acid observed after a four-week treatment-free recovery period, the applicant clarifies that the mean bile acid level in females was slightly higher than in the concurrent controls, but this was attributed to the lower values in the controls rather than to an actual finding related to the test object. Overall, the liver findings were classified non-adverse and of benign nature.

Several other clinical observations, plasma chemistry, gross or microscopic findings were noted in the repeated dose toxicology programme. However, they were considered to be non-test article related, transient, sporadic, attributable to the palatability of the test article, no longer present at the end of the recovery period and covered by adequate safety margins

Steady state total/free AUC multiples relative to MRHD at the NOAEL at the toxicology studies are 28/13 (1-month toxicity study in rats NOAEL: 15 mg/kg), 14/33 (1-month toxicity study in dogs; NOAEL: 15 mg/kg), 20/10 (6-month toxicity study in rats; NOAEL: 9 mg/kg), 1/3 (6-months toxicity study in dogs; NOAEL: 2 mg/kg) and 5/12 (9-month study in dogs; NOAEL: 8 mg/kg).

Overall, the exposure ranges obtained in the repeated dose toxicity studies are considered acceptable to ensure adequate clinical safety. Effects in non-clinical studies were observed only at exposures considered sufficiently in excess of the maximum human exposure indicating little relevance to clinical use as stated in section 5.3 of the SmPC.

Genotoxicity

Based on the results of a GLP-conform standard battery of *in vitro* and *in vivo* genotoxicity tests in line with ICH S2(R1) brensocatib can be considered as non-genotoxic.

Carcinogenicity

No neoplastic findings were observed in a 6-month Tg Ras mouse study and a 104-week carcinogenicity study in rats up to the highest doses tested yielding AUC-based (unbound brensocatib fraction) safety margins of > 60-fold (mice) and > 50-fold (rat) to the MRHD. Therefore, no carcinogenic risk is anticipated for patients.

Developmental and reproductive toxicity

A complete evaluation of reproductive and developmental toxicity of brensocatib has been performed in line with the ICH S5 (R3) guideline.

Although no dedicated studies on fertility and early embryonic development (FEED) have been performed, these evaluations were included in the 6-months repeat-dose toxicity study in rats for male fertility and a definitive combined fertility and embryo-foetal developmental study in rats for female fertility. This is acceptable.

Brensocatib had no effects on rat male and female fertility, mating performance and female estrous cycles.

The NOAEL for male fertility and female fertility were set at 50 mg/kg/day and 100 mg/kg/day, respectively. The safety margins to human exposure at the maximum recommended dose of 25 mg/day of brensocatib based on the AUC is 150-fold/75-fold for total/free exposure and 128/61-fold for total/free exposure for male and female fertility, respectively.

Potential effects of brensocatib on embryo-foetal development were investigated in rats and rabbits in dose-range findings and definitive EFD studies.

In the **definitive fertility and embryo-foetal developmental study** in rats administration of brensocatib resulted in slight maternal toxicity (e.g. changes in body weight gain and food consumption) at 20 or 100 mg/kg/day at the first days of treatment, only. Whereas no major malformations or external or visceral abnormalities/variations were observed, minor skeletal malformations of bent scapula and wavy ribs were noted after dosing with 100 mg/kg/day. There was an increased incidence of skeletal variations (malpositioned pelvic girdle and vestigial supernumerary full and/or short ribs in both cervical and thoracolumbar regions, incomplete ossification of sternebra) and differences in ossification after dosing with 20 or 100 mg/kg/day. After dosing with 3 mg/kg/day, there was reduced incidence of ossification in one skull bone only. An increased incidence of ossification of the phalanges (both forepaw and hindpaw) was seen at all dose levels of the hindlimb calcaneous at 20 or 100 mg/kg/day and of the interparietal skull at 100 mg/kg/day, indicating more advanced ossification in some fetuses in brensocatib-treated groups. The NOAEL for maternal toxicity and EFD was considered to be 3 mg/kg/day corresponding to a systemic exposure of 2.73 µM and 28.7 µM*h for C_{max} and $AUC_{0\text{-}24h}$ values on GD 16, respectively. The AUC-based safety margin to human exposure at the maximum recommended human dose of 25 mg/day is only 3-fold for total exposure and 1-fold for free exposure.

According to the ICH S5(R3) guideline, if the NOAEL occurs at an exposure margin <10-fold, findings are of increased concern.

The applicant discussed that the skeletal findings in rat foetuses are unlikely to result from the slight maternal toxicity which occurred in the in the premating phase, only.

According to the applicant, the skeletal findings of wavy ribs and bent scapulae should not be used in risk assessment to set safe doses or safety margins, since these findings should be considered as variations rather than malformations and have been shown to be transient and reversible postnatally in rats. The view of the applicant that these findings should rather be considered as variations or minor malformations (in case of bent scapula) is agreed and supported by the literature (Baier et al., 2016, Kimmel, 2014; Roos et al., 2024; Mitchard, 2014).

However, it is not supported that these findings should not be used for risk assessment. Wavy ribs and bent scapulae occurred in the absence of maternal toxicity and fetal weight reduction in a clear dose-dependent manner and at incidences above the historical control. In addition, further skeletal variations above the incidences of historical controls (malpositioned pelvic girdle and vestigial supernumerary full and/or short ribs in both cervical and thoracolumbar regions) were observed in a dose-dependent manner. Also, there were changes in ossification at all dose levels: the incidence of foetuses with

ossification of the phalanges (both forepaws and hindpaws) was increased above control levels at all brensocatib dose levels and at 20 or 100 mg/kg/day increased ossification of the hindlimb calcaneous was observed. These findings indicate that ossification was more advanced in a greater number of foetuses in brensocatib-treated groups, than in controls. At 100 mg/kg/day, the incidence of minimal incomplete ossification of the interparietal skull was lower than control levels, and there was increased incidence of the vertebrae cervical centrum unossified. The incidence of incomplete ossification of the parietal skull bone was increased dose levels.

As discussed by Carney and Kimmel, 2007, when delayed ossification or associated findings like wavy ribs and bent scapula occur in the absence of maternal toxicity one must consider the possibility of a more specific effect on fetal ossification or chondrogenesis.

Therefore, a direct substance-related effect cannot be excluded and the relevance for humans is currently unknown. SmPC section 5.3 adequately reflects current nonclinical findings.

The 3 mg/kg/day as defined in the study report should be considered as the NOAEL for rat embryo-foetal development.

In the definitive EFD study in rabbits there was slight maternal toxicity (reduction in body weight gain and food consumption) at 50 mg/kg/day only. There were no brensocatib-related effects on embryo-foetal development and survival. The increase in several ossification parameters in the brensocatib-administered groups (unossified/incompletely ossified odontoid process, cervical vertebral centrum/a and cervical vertebral arch(es) at 15 and 50 mg/kg/day along with cranial bone(s) small linear/linear ossification irregularity/ies and sacral vertebral centrum connected to sacro-iliac articulation centre at 50 mg/kg/day only) lay within the historical control data and are therefore not related to brenosocatib treatment. An increased incidence of pelvic girdle caudal displacement associated with supernumerary ribs on the 13th thoracic vertebra occured at 15 and 50 mg/kg/day. Although the applicant discussed that there is a potential test item-relation for the supernumerary 13th ribs and pelvic girdle caudal displacement, these findings are common background findings in rabbits and the absence of major skeletal malformations support the conclusion that no adverse skeletal findings were observed in the rabbit EFD study following administration with brensocatib. However, the incidences of supernumerary 13th ribs and pelvic girdle caudal displacement, are above the historical controls of the last 5th year period and similar findings have been observed in rats. But in contrast to rats, for rabbits an influence of maternal toxicity is possible. Therefore, the increased incidence of supernumerary ribs on the 13th thoracic vertebra and pelvic girdle caudal displacement at 15 and 50 mg/kg/day in association with slight maternal toxicity at these dose levels can be viewed as non-adverse and the NOAEL for developmental toxicity in rabbits of 50 mg/kg/day is agreed.

In the prenatal- and postnatal developmental study in rats brensocatib up to the highest dose level did not induce maternal toxic effects nor effects on pre- and postnatal development. The safety margin to the maximum recommended human exposure at 25 mg/day at the NOAEL of 20 mg/kg/day is 17/8-fold for total/free exposures. Embryofoaetal toxicity is reflected in RMP as an important postential risk.

Data from the bioanalysis of brensocatib in plasma of pups on Lactation/Postnatal Day (LD/PND) 4 showed exposure of the pups at all dose levels which likely occur via breast milk.

Studies in juvenile animals are not warranted for treatment in patients 12 years of age and older since the repeat-dose toxicity studies in dogs and rats cover the age of the intended paediatric patient population.

Other toxicity studies

Immunotoxicity: No dedicated immunotoxicity investigations were conducted and there was no evidence of a concerning immunotoxicological risk based on a comprehensive weight-of-evidence evaluation in

accordance with ICH S8 guideline.

Although some immune-related changes were observed in the nonclinical toxicity studies in rats and/or dogs (histological changes was the observation of PLD in the lymphoid tissues and periodontitis in dogs) they were reversible, dose-dependent, and occurred primarily at the highest dose evaluated in each study. In the completed clinical programme, there have been no indications of brensocatib-related increases in the incidence of adverse events related to immunotoxicity or autoimmunity. Therefore, no additional nonclinical testing for immunotoxicity was considered necessary. The non-clinical findings are reflected in section 5.3 of the SmPC. It can be agreed that further specific immunotoxicity studies are not warranted.

Dependency: No dedicated studies for assessing abuse/dependence were conducted and there was no evidence of such a risk based on a comprehensive weight-of-evidence evaluation. The risk of dependence is considered low and therefore specific dependency studies are not warranted.

Ecotoxicity/environmental risk: Brensocatib is not a PBT substance. Considering the available data, brensocatib is not expected to pose a risk to the environment.

Studies on Metabolites: Based on the available data, no additional non-clinical studies to assess the toxicity of are considered necessary to qualify the only major human metabolite, M8 (thiocyanate). There are no other metabolites of concern.

Studies on Impurities: The nitrosamine impurity, was identified. The nitroso impurity was negative in an EAT supported by a negative in vitro MLA performed with rat S9 mix, human metabolism data, which indicate a low potential for α-hydroxlation, and quantum mechanic modelling supportive of low carcinoghenic potency of the nitroso impurity. Overall, the weight of evidence indicates a low mutagenic potential of the nitroso impurity and is also supported by NSOEG.

Other mutagenic impurities are adequately controlled in accordance with ICH M7(R2). The non-mutagenic impurities are considered toxicologically qualified above the qualification threshold in accordance with ICH Q3 guidance.

Phototoxicity: Brensocatib was not phototoxic according to the ICH guideline S10. The lack of stand-alone toxicological studies on local tolerance, antigenicity, immunotoxicity, dependence potential, metabolites, (conventional) impurities or excipients was adequately justified so that no further testing is required.

Excipient studies: There are no novel excipients.

5.5.2. Conclusion

The nonclinical data are adequate to support an approval. The animal toxicity studies findings have been adequately described in the SmPC section 5.3.

The findings of the toxicity, developmental and reproductive toxicity studies are adequately reflected in sections 4.6 and 5.3 of the SmPC.

6. Clinical aspects

6.1. Introduction

6.1.1. Good Clinical Practice (GCP) aspects

The clinical trials were performed in accordance with GCP as claimed by the applicant.

The applicant has provided a statement to the effect that clinical trials conducted outside the community were carried out in accordance with the ethical standards of Directive 2001/20/EC.

Based on the review od the clinical data, CHMP did not identify the need for a triggered GCP inspection.

6.1.2. Tabular overview of clinical trials

Table 5: Tabular overview of main clinical studies

Study ID	Design, control type, duration	Treatment	Subject population	Study objectives and primary endpoint	Number of subjects total and per group randomised (treated)/completed study
Phase 2					
INS1007-201 (WILLOW)	RD, DB, PC, 24 weeks	Brensocatib 10 or 25 mg QD, placebo	Adults with NCFBE	Efficacy, safety; Primary: Time to first PE	256 / 256 / 225
Phase 3					
INS1007-301 (ASPEN)	RD, DB, PC, 52 weeks	Brensocatib 10 or 25 mg QD, placebo	Adults and adolescents with NCFBE	Efficacy, safety; Primary: Annualized rate of PEs	1721 / 1721 / ~1600

RD = randomised; DB = double blind; PC = placebo controlled; QD = once daily; PE = Pulmonary exacerbation

6.2. Clinical pharmacology

6.2.1. Methods

Bioanalytical methods

Two bioanalytical methods for quantification of brensocatib (also known as AZD7986 and INS1007) in plasma and one bioanalytical method for quantification of brensocatib in urine were established in different laboratories. All LC-MS/MS methods were fully validated and cross-validated to ensure consistency of concentration data.

Table 6: Pharmacokinetic Assays

Analyte	Bioanalytical Study Number	Matrix	Bioanalytical Site	Analytical Range (ng/mL)	LLOQ (ng/mL)	QC Performance: %bias	QC Performance: CV%	Clinical Study Number
AZD7986 (brensocatib)	8309775	K_2EDTA Plasma	Labcorp Harrogate, UK	0.841 to 841 (2 to 2000 nmol/L)	0.841	Intra-assay: -8.0 to -6.5% (LLOQ), -5.6 to 3.3% (above LLOQ); Inter-assay: -7.5% (LLOQ), -3.8 to 1.0% (above LLOQ)	Intra-assay: 4.6 to 8.4% CV (LLOQ), 1.0 to 5.3% CV (above LLOQ); Inter-assay: 6.2% CV (LLOQ), 2.5 to 4.1% CV (above LLOQ)	D6190C00001 D6190C00003
INS1007 (brensocatib)	VISMN1700 P1	K_2EDTA Plasma	KCAS, USA	0.250 to 150	0.250	Intra-assay: -4.0 to 2.4% (LLOQ), -4.1 to 0.8% (above LLOQ); Inter-assay: -1.4% (LLOQ), -2.7 to 0.3% (above LLOQ)	Intra-assay: 4.5 to 6.2% CV (LLOQ), 1.4 to 7.3% CV (above LLOQ); Inter-assay: 5.9% CV (LLOQ), 2.0 to 5.7% CV (above LLOQ)	INS1007-101 INS1007-102 INS1007-201 INS1007-211 INS1007-301
INS1007 (brensocatib)	8462136	K_2EDTA Plasma	Labcorp Madison, USA	0.841 to 841	0.841	Intra-assay: -2.0% (LLOQ), -4.5 to 2.1% (above LLOQ)	Intra-assay: 5.1% CV (LLOQ), 2.1 to 4.7% CV (above LLOQ)	INS1007-103 INS1007-104 INS1007-105 INS1007-106 INS1007-109
Analyte	**Bioanalytical Study Number**	**Matrix**	**Bioanalytical Site**	**Analytical Range (ng/mL)**	**LLOQ (ng/mL)**	**QC Performance: %bias**	**QC Performance: CV%**	**Clinical Study Number**
AZD7986 (brensocatib)	8309776	Urine	Labcorp Harrogate, UK	42.0 to 42000 (100 to 100000 nmol/L)	42.0	Intra-assay: -5.6 to -1.0% (LLOQ), -3.9 to 5.8% (above LLOQ); Inter-assay: -4.4% bias (LLOQ), -2.5 to 3.8% (above LLOQ)	Intra-assay: 3.5 to 7.4% CV (LLOQ), 1.2 to 4.8% CV (above LLOQ); Inter-assay: 6.8% CV (LLOQ), 2.9 to 3.6% CV (above LLOQ)	D6190C00001
INS1007 (brensocatib)	8462135	Urine	Labcorp Madison, USA	42.0 to 42000	42.0	Intra-assay: -5.2% (LLOQ), -0.6 to 9.0% (above LLOQ) Additional Intra-assay (Amendment 1): -3.0 to 8.7 % bias	Intra-assay: 7.1% CV (LLOQ), 2.3 to 5.5% CV (above LLOQ)	INS1007-103 INS1007-105

Source: Section 4.1
LLOQ = lower limit of quantification, QC = quality control, CV = coefficient of variation, K_2EDTA = potassium salt of ethylenediaminetetraacetic acid (anticoagulant).

An exploratory LC-MS/MS method was developed and qualified for analysis of thiocyanate, the only major metabolite identified for brensocatib, in early clinical studies. The method was subsequently validated and used for monitoring of potential thiocyanate-related adverse events in patients with potentially associated adverse events in Phase 2 and Phase 3 studies.

Table 7: Metabolite Assays

Analyte	Bioanalytical Study Number	Matrix	Bioanalytical Site	Analytical Range (µg/mL)	LLOQ (µg /mL)	QC Performance: %bias	QC Performance: CV%	Clinical Study Number
Thiocyanate	2206017	K_2EDTA Plasma	Resolian, USA	Original range: 0.500 to 30.0; Final range: 0.200 to 20.0	0.200	Artificial plasma: 94.0 to 106.8% accuracy (original range), 96.1 to 100.6% accuracy (final range); Human plasma: 99.3 to 109.7% accuracy (original range), 98.9 to 104.6% accuracy (final range)	Artificial plasma: ≤ 5.7% CV (original range), ≤ 2.76% CV (final range); Human plasma: ≤ 2.6% CV (original range), ≤ 1.86% CV (final range)	INS1007-102 INS1007-104 INS1007-105 INS1007-201
Thiocyanate	V2304024	K_2EDTA Plasma	Resolian, USA	0.200 to 20.0	0.200	0.9 to 9.7% bias (LLOQ-QC, LQC in artificial human plasma; MQC, HQC in human plasma)	≤ 6.8% CV (LLOQ-QC, LQC in artificial human plasma; MQC, HQC in human plasma)	INS1007-301

Source: Section 4.2
CV = coefficient of variation, HQC = high quality control, K_2EDTA = potassium salt of ethylenediaminetetraacetic acid (anticoagulant), LLOQ = lower limit of quantification, LQC = low quality control, MQC = medium quality control, QC = quality control.

PD biomarkers

Different bioanalytical methods for the quantification of NSPs were used throughout clinical studies. A semi-quantitative validated bioanalytical method was used to analyze the activity of NE in plasma in Study D6190C00001. In this assay, unstimulated (saline) or stimulated (zymosan) plasma samples were added to a 96-well plate containing a fluorogenic substrate cleaved by NE. The amounts of the fluorescent product were measured to determine the relative activity of NE in the samples. Stimulated samples in the presence of an NE inhibitor served as the positive control.

Concentrations of active NE, PR3, and CatG in sputum were determined in Study INS1007-201 using qualified kit-based colorimetric enzyme-linked immunosorbent assay (ELISA) assays. For Studies INS1007-211 and INS1007-301, kinetic enzyme activity assays were used to quantify the concentration of active NE, PR3, and CatG in both sputum and WBC pellets. (For summarized overview, see 1.2.2, Mod.2.7.1).

6.2.2. Pharmacokinetics

6.2.2.1. Introduction

Brensocatib is proposed to be administered as 25 mg film-coated tablet to be taken once daily irrespective of meals. A clinical pharmacology data package was provided encompassing ADME data after administration of ^{14}C brensocatib, SAD / MAD PK characterization in healthy volunteers, examination of food effect, intrinsic factors like exposure in renal and hepatic impairment, extrinsic factors like DDI (incl. different CYP3A4 and P-gp inhibitors like verapamil, itraconazole, clarithromycin; CYP3A4 inducer rifampin; and PPI esomepromazole), secondary pharmacology (QT/QTc prolongation), and PD biomarker measurement of target enzyme NSP activity in plasma and sputum.

6.2.2.2. Evaluation and qualification of models

6.2.2.2.1. Population pharmacokinetics

Population PK model

Several reports were provided to support the PK model.

Report ICPD 00566-1

The objectives of these analyses were the following:

To develop a population pharmacokinetic (PK) model characterizing the disposition of INS1007 in plasma using data collected from subjects enrolled in Studies INS1007-101 and INS1007-201;
To explore PK-PD relationships for efficacy and safety of INS1007 in subjects enrolled in INS1007-201; and
To assist in the selection of doses for future clinical INS1007 studies using model-based simulations.

The population PK analysis was conducted using NONMEM® software Version 7.2 (ICON Development Solutions, Ellicott City, MD) implementing the first-order conditional estimation method with eta-epsilon (η-ε) interaction.

Base structural model development was conducted using the intensive PK sampling data from INS1007-101. The base structural model was then applied to a pooled dataset containing INS1007-101 data and that from INS1007-201 and refined as necessary.

The full population PK dataset, pooled from INS1007-101 and INS1007-201, was comprised of 226 individuals and 2428 plasma samples. The PK data obtained after the first dose (n = 1,144) had to be excluded to allow for co-modelling of data across studies.

Table 8: Description of data included in population PK analyses

Study	# of Subjects/Samples Available at the start of analysis		# of Subjects/Samples from Placebo, Subjects		# of other Samples Excluded		Current # of Subjects/Samples available for analysis	
INS1007-101	82/2344		13/308		171[a]		69/1865	
INS1007-201	234/1002		76/315		122[b]		158/563[c]	
	Intense	Sparse	Intense	Sparse	Intense	Sparse	Intense	Sparse
	29/490	205/512	9/153	67/162	98	24	20/238a	138/325[c]

Note: Abbreviations are provided in the Abbreviation Listing.
a. Includes: 7 BLQ post-dose samples; 164 records prior to the first dose that were BLQ and removed
b. Includes 76 post-dose BLQ samples; 5 samples with very long time since dose (inconsistent with sampling scheme); 11 samples removed for lack of information on previous dose; 21 samples with issues with irreconcilable issues with dose date/time; 9 outliers
c. There were two duplicate records sampled at same time but having different concentrations, occurring for two subjects. So average concentration was calculated at that sample and one of the records for each subject was removed, as NONMEM cannot handle duplicate times.

Thus, the final dataset for population PK model development included steady-state data from 225 subjects who contributed 1284 INS1007 concentrations. The overall pooled analysis population was 36.9% male, with broad ranges of age (20 to 83 years), weight (40.4 to 155 kg), and renal function (26.7 to 149 mL/min/1.73 m2). Patients enrolled in INS1007-201 tended to be older and have lower renal function, though variability in demography was of similar extent as in subjects from INS1007-101.

The final population PK model, which was fit to the pooled data from both studies, included two distribution compartments, linear elimination, and a 6-chain transit compartment to account for the variability observed in the rate of absorption; separate absorption rate constants were used for the fed and fasted state. Residual variability (RV) was described using a combined additive plus proportional error model. In order to allow for co-modelling of phase 1 and Phase 2 data, body weight scaling was employed prior to the conduct of the covariate analysis.

A formal covariate analysis was conducted using stepwise forward inclusion ($p < 0.05$, $\Delta MVOF \geq 3.84$ units) and backward elimination ($p < 0.001$, $\Delta MVOF \geq 10.8$ units). This was conducted using the automated stepwise covariate modeling (scm) algorithm implemented in Perl-Speaks NONMEM (PSN). Covariate analyses resulted in the identification of two statistically significant relationships: 1) between apparent oral volume of the central compartment (Vc/F) and subject age, and 2) between apparent oral clearance (CL/F) and subject renal function [estimated using creatinine clearance (CLcr)].

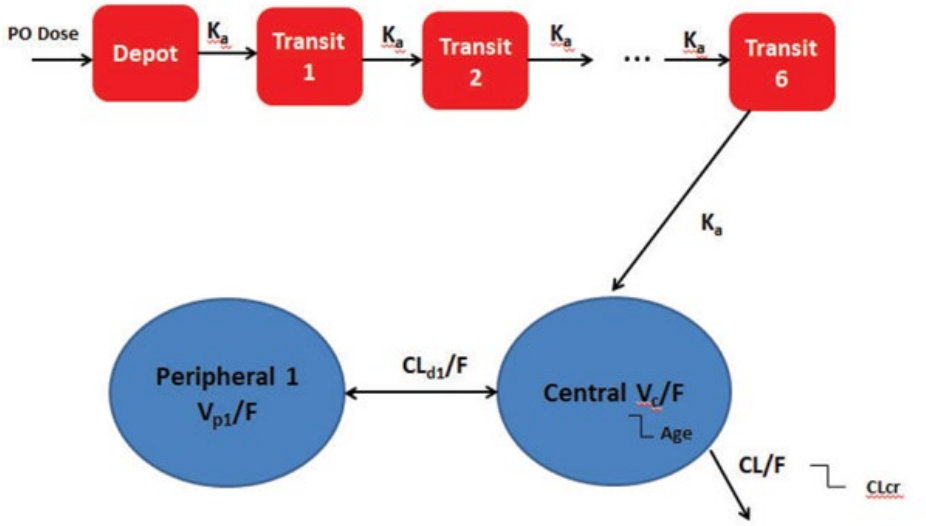
The Bayesian PK parameter estimates obtained from the fit of the final population PK model to the pooled steady-state data were used to generate individual predicted steady-state concentration-time profiles for each patient. The peak plasma concentration (Cmax) at steady-state was determined by direct observation using the individual predicted concentrations. The area under the plasma concentration-time curve from 0 to 24 hours (AUC0-24) at steady-state was calculated by integrating the individual predicted concentration-time profile over 24 hours at steady-state.

Table 9: Population pharmacokinetic parameter estimates for the final model (pooled Phase 1 and 2, steady-state data only)

Parameter	Population Mean		Magnitude of IIV (CV%)		
	Estimate	%SEM	Estimate	%SEM	%Shrinkage
CL/F (L/h/70kg)	6.23	0.208	37.3	5.38	< 1.0
Vc/F (L/70kg)	196	0.208	43.9	1.16	36.6
CLd/F (L/h)	6.26	0.202	---	---	---
Vp/F (L/70kg)	80.6	0.266	---	---	---
K_{aFast} (1/h)	11.7	0.292	30.4	8.97	31.2
K_{aFed} (1/h)	9.96	0.577			
Vc/F:Age (power)	0.396	13.1	---	---	
CL/F: CLcr (power)	0.276	12.0	---	---	
Proportional RV (%CV)	15.9	2.01	---	---	5.17
Additive RV (ng/mL)	0.000119	6.72	---	---	
			Correlation coefficient	r^2	
Cov(IIV_CL,IIV_V_C) (CV%)	21.5	14.3	0.283	0.0801	
Condition number: 9467.7					

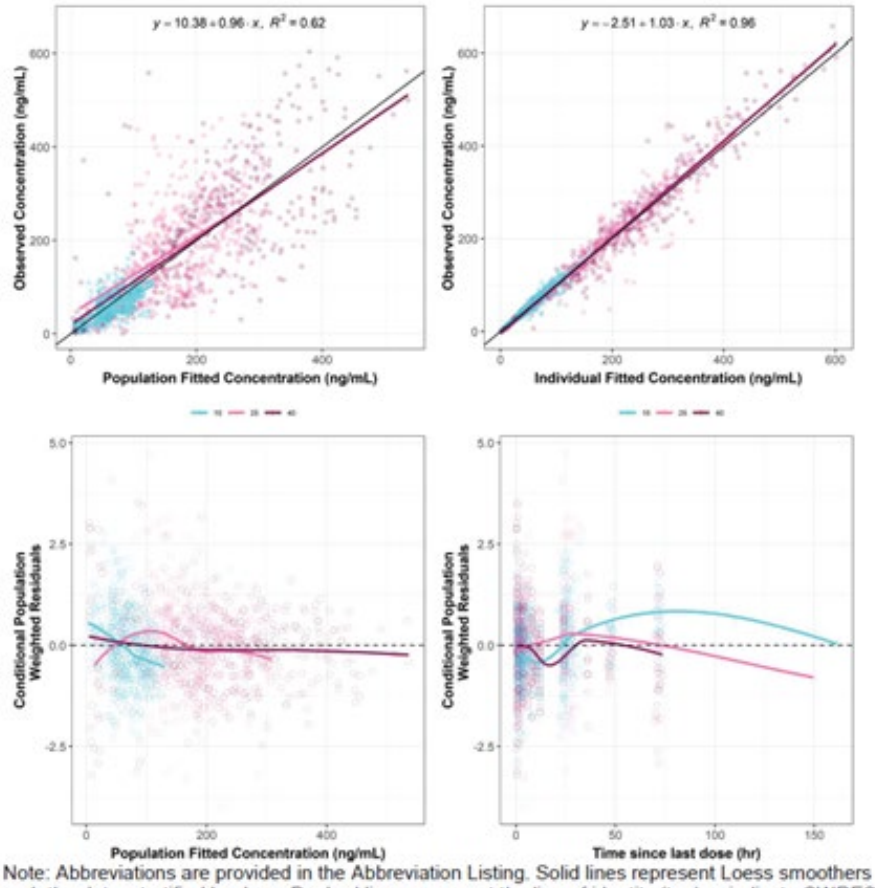
Note: Abbreviations are provided in the Abbreviation Listing.
Note: To facilitate co-modeling of data from Phase 1 and 2, clearance and volume parameters were allometrically scaled *a priori* using fixed exponents of 0.75 and 1, respectively. CLd/F was not scaled as the model failed to converge when using allometric scaling for that parameter.

Figure 2: Schematic of final pooled population PK model



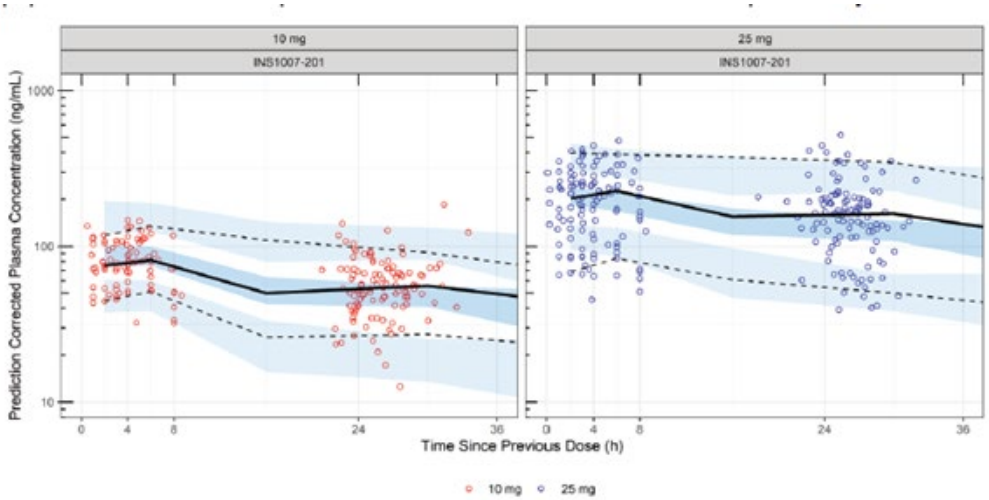
Note: Abbreviations are provided in the Abbreviation Listing. K_a in the above diagram represents both K_{aFast} and K_{aFed}, which are invoked based upon whether or not the dose was given in the fasted state

Figure 3: Standard goodness-of-fit plots for the final model (pooled Phase 1 and 2,steady-state data only), colored by INS1007 dose



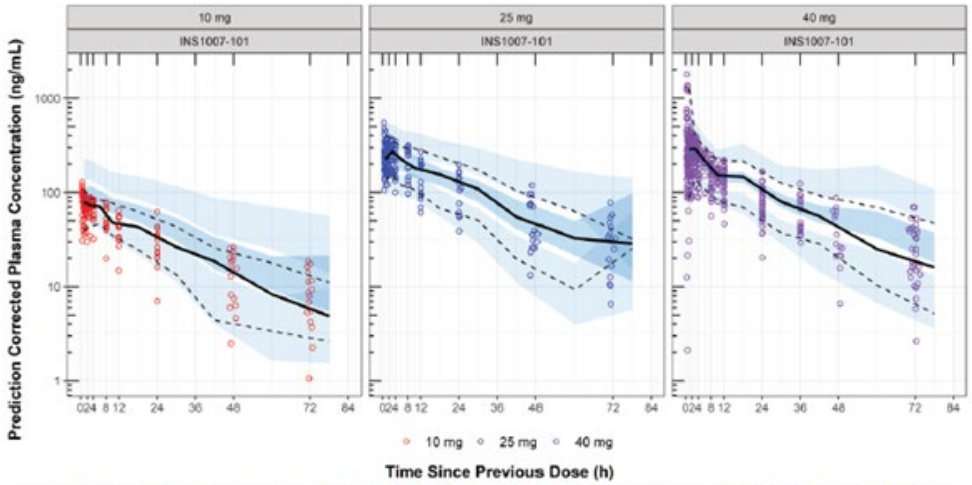
Note: Abbreviations are provided in the Abbreviation Listing. Solid lines represent Loess smoothers through the data, stratified by dose. Dashed lines represent the line of identity (top) or indicate CWRES=0 (bottom).

Figure 4: Prediction-corrected visual predictive check plots for the final pooled population PK model compared to observed data from INS1007-201, paneled by dose



Note: Abbreviations are provided in the Abbreviation Listing. Open circles are observed concentrations, black solid lines are the median observed concentrations, black dashed lines are the 5th and 95th percentiles of the observed concentrations. Blue shaded regions are the 90% confidence intervals for the median, 5th, and 95th percentiles from the simulations.

Figure 5: Prediction-corrected visual predictive check plots for the final pooled population PK model compared to observed data from INS1007-101, paneled by dose



Note: Abbreviations are provided in the Abbreviation Listing. Open circles are observed concentrations, black solid lines are the median observed concentrations, black dashed lines are the 5th and 95th percentiles of the observed concentrations. Blue shaded regions are the 90% confidence intervals for the median, 5th, and 95th percentiles from the simulations.

Report Number: ICPD 00694-1

The objectives of these analyses were the following:

To construct a population PK model characterizing the disposition of brensocatib; and,

To explore E-R relationships for efficacy and safety of brensocatib in patients enrolled in INS1007-201 and INS1007-301; and

To conduct model-based simulations to explore potential differences in predicted outcomes base on brensocatib dose.

The population PK analysis was conducted using NONMEM® version 7.4.4 (ICON Development Solutions, Ellicott City, MD) implementing the first-order conditional estimation method with η-ε interaction.

The previously developed population PK models for brensocatib were used to inform structural model development for the population PK model and were refined, as necessary, using the full pooled dataset. The full pooled dataset used in the analyses reported herein consisted of 11 clinical trials (eight Phase 1, two Phase 2, and one Phase 3).

The Bayesian PK parameter estimates obtained from the fit of the final population PK model to the pooled data were used to generate individual predicted steady-state concentration-time profiles for each subject, which were then used to estimate brensocatib exposure parameters in each subject (area under the concentration-time curve from 0 to 24 hours at steady-state [AUC24], steady-state maximum plasma concentration [Cmax], and steady-state minimum plasma concentration [Cmin]).

A dataset of 11,643 brensocatib plasma concentration observations collected from 1,098 healthy volunteers (n = 291), patients with cystic fibrosis (CF) (n = 24), and patients with NCFBE (n = 783) was used for the construction of the population PK model. The final population PK model for brensocatib that best described the observed data consisted of two systemic compartments with linear elimination to describe the overall drug disposition and an oral depot compartment with a 3-compartment transit chain followed by first-order absorption. The overall pooled analysis population (n = 1,098) was 55.4% female, with broad ranges of age (12 to 85 years), weight (31.7 to 155.49 kg), and renal function (12.89 to 204.2 mL/min/1.73 m2).

Interindividual variability was estimated for the absorption rate constant (Ka), apparent clearance (CL/F), apparent volume of the central compartment (Vc/F), apparent volume of the peripheral compartment (Vp/F), and relative bioavailability term in the population PK model (relF) while interoccasion variability (IOV) was estimated for Ka and relF. Residual variability was described using an additive plus proportional error model distinguished by study phase. covariate model development was undertaken using forward selection followed by a backward elimination procedure. This was conducted using the automated stepwise covariate modeling algorithm implemented in Perl-speaks NONMEM.

Variability in the population PK parameters was found to be related to the following factors:

- CL/F – significantly related to hepatic impairment, weight, sex (female), race (Black or Asian), and concomitant medications (rifampin, clarithromycin, or verapamil).
- Vc/F – significantly related to age and weight.
- Q/F – significantly related to weight.
- Vp/F – significantly related to weight.
- Ka – significantly related to fed status and oral solution formulation.
- relF – significantly related to dose and concomitant medications (clarithromycin or verapamil). Relative bioavailability was found to be dosedependent and explained well by a standard sigmoidal maximum obtainable percent change from baseline (Emax) function

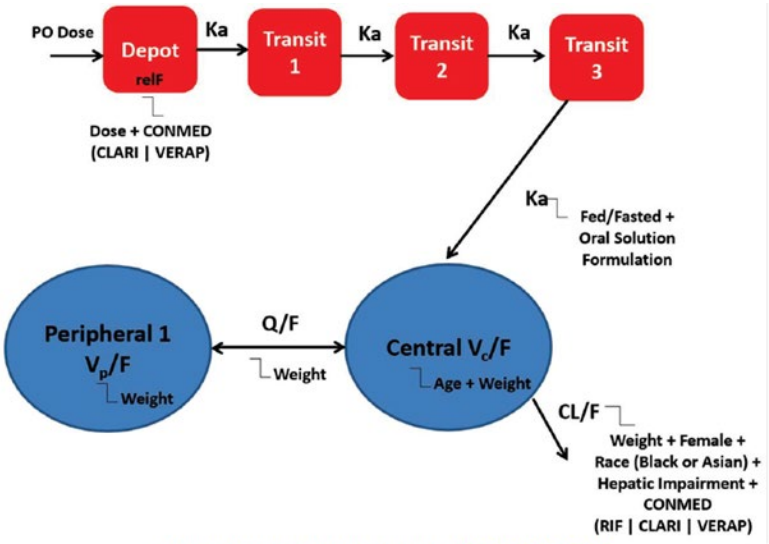
Despite statistical significance, the magnitude of effect of these covariates was relatively modest. Neither renal function nor disease status (i.e., indication of healthy, CF, or NCFBE) were found to be significant predictors of the variability in brensocatib PK.

The previous population PK model was used as the base model for fitting the pooled dataset of 11 Phase 1, 2, and 3 brensocatib studies. The population PK model developed deemed to best represent the system was a simple 2- compartment mammillary model with absorption described via a depot compartment followed by 3 transit compartments. The base model covariate relationships included CLcr and weight on CL/F, age and weight on the apparent volume of the central compartment (Vc/F), weight on the volume of the peripheral compartment (Vp/F), fed status on the absorption rate constant (Ka). The

residual variability model found to best represent the system consisted of additive plus proportional error terms.

There was a statistically significant improvement in the model fit when accounting for the IIV in CL/F and relF relating to concomitant medication use. The IIV in CL/F relating to concomitant medication use was found to be significant for rifampin, clarithromycin, and verapamil whereas only clarithromycin and verapamil were significant descriptors of IIV in relF.

Figure 6: Schematic of final pooled population PK model



Note: Abbreviations are provided in the Abbreviation Listing.

Table 10: Summary statistics of resampled population PK parameters in comparison to the fitted population PK model parameter estimates (population mean parameters)

Parameter	Final model			Resample statistics (N = 1000)			
	Final estimate	%SEM	Shrinkage	Mean	Median	%CV	90% CI
CL/F (L/hr/70kg)[a]	3.25	0.0838	2.58	3.2	3.19	2.28	[3.08, 3.32]
CL/F-Hepatic Impairment[b]	0.187						
CL/F-Rifampin[c]	0.383						
CL/F-Clarithromycin[d]	-0.0779						
CL/F-Verapamil[e]	0.212						
CL/F-Female	-0.0957	0.0235	24.5	-0.0966	-0.0968	24.7	[-0.135, -0.0580]
CL/F-Black	0.492	0.0791	16.1	0.557	0.551	12.5	[0.448, 0.672]
CL/F-Asian	0.229	0.0442	19.3	0.216	0.219	20.1	[0.142, 0.285]
Vc/F (L/70kg)[a]	78.8	1.45	1.84	79.5	79.4	2.05	[76.9, 82.3]
Vc/F-Age	0.314	0.0312	9.94	0.27	0.269	11.3	[0.221, 0.319]
Q/F (L/hr/70kg)[a]	5.97	0.167	2.8	5.92	5.92	2.78	[5.65, 6.20]
Vp/F (L/70kg)[a]	50.4	0.87	1.72	52.9	52.9	1.76	[51.4, 54.4]
Ka-Fasted (1/hr)	7.61	0.236	3.1	7.43	7.45	3.07	[7.04, 7.79]
Ka-Fed (1/hr)	5.8	0.415	7.16	4.62	4.62	7.93	[4.02, 5.19]
Ka-Oral Solution Formulation	0.809	0.199	24.6	0.911	0.905	17.3	[0.652, 1.17]
E0	0.29	0.011	3.8	0.284	0.283	3.75	[0.267, 0.302]
Emax	0.346	0.0221	6.39	0.332	0.332	7.61	[0.291, 0.373]
EC50 (mg)	21.6	1.36	6.29	17.1	17.1	11.5	[13.9, 20.1]
H	1.54	0.0928	6.03	1.31	1.31	6.57	[1.17, 1.45]
relF-Clarithromycin[d]	0.388						
relF-Verapamil[e]	0.657						

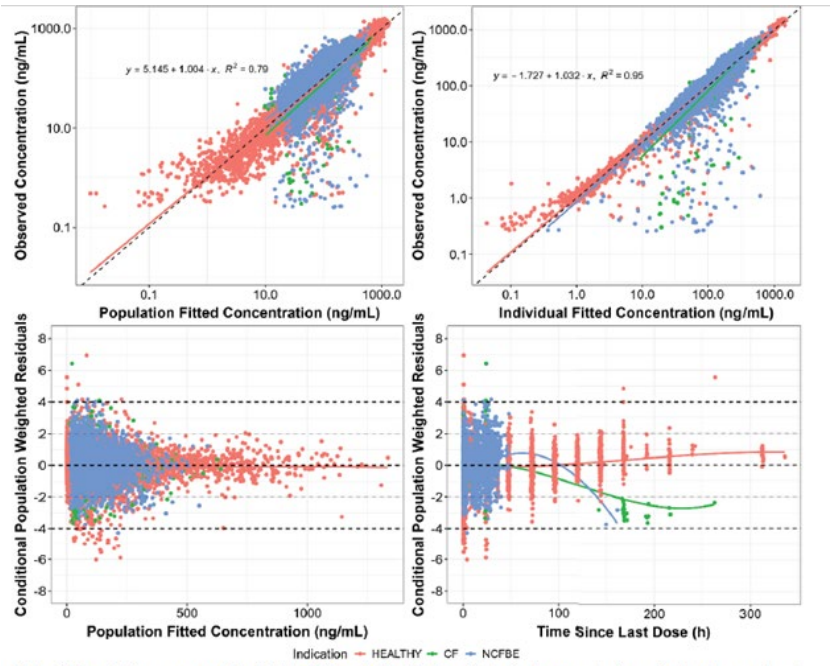
Note: Abbreviations are provided in the Abbreviation Listing.
a. Clearance and volume parameters were allometrically scaled using fixed exponents of 0.75 and 1, respectively.
b. Entirely informed by data from Study INS1007-105.
c. Entirely informed by data from Study INS1007-106.
d. Entirely informed by data from Study INS1007-109.
e. Entirely informed by data from Study D6190C00003.

Table 11: Summary statistics of resampled population PK parameters in comparison to the fitted population PK model parameter estimates (population mean parameters)

Parameter	Final model			Resample statistics (N = 1000)			
	Final estimate[c]	%SEM	Shrinkage	Mean	Median	%CV	90% CI
IIV CL/F	0.12 (34.7%)	6.53	19.1	0.121	0.121	5.8	[0.108, 0.132]
IIV Vc/F	0.0494 (22.2%)	11.1	43.9	0.0581	0.058	8.61	[0.0496, 0.0659]
IIV Vp/F	0						
IIV Ka	0.0233 (15.3%)	18	63.8	0.0336	0.0335	13.7	[0.0264, 0.0419]
IIV relF	0.164 (11.5%)	18.6	58.1	0.128	0.128	22.4	[0.0825, 0.174]
IOV Ka[a]	0.0132 (42.9%)	36.7	64.5	0.0214	0.0215	18.5	[0.0143, 0.0281]
IOV relF[b]	0.184 (18.8%)	10.4	56	0.213	0.213	7.86	[0.184, 0.240]
Phase 1 Proportional RV (%CV)	0.0353	9.65	50.1	0.0189	0.0188	10.6	[0.0158, 0.0223]
Phase 1 Additive RV (ng/mL)	0.0244	0.643	9.2	0.0239	0.0239	0.823	[0.0236, 0.0243]
Phase 2 Proportional RV (%CV)	0.0919	5.79	9.2	0.0883	0.0875	7.19	[0.0795, 0.100]
Phase 2 Additive RV (ng/mL)	0.0647	1.12	1E-10	0.066	0.066	1.09	[0.0648, 0.0672]
Phase 3 Proportional RV (%CV)	0.407	0.111	1E-10	0.408	0.408	0.111	[0.407, 0.409]
Phase 3 Additive RV (ng/mL)	0.119	1.94	11.9	0.119	0.12	2.29	[0.115, 0.124]

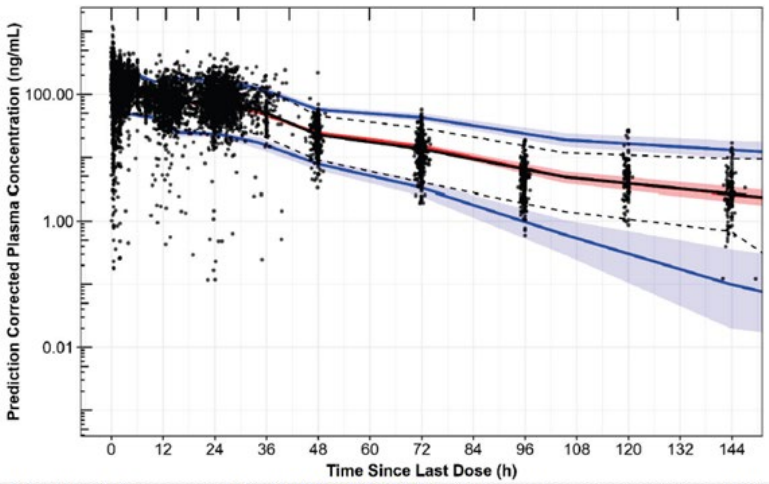
Note: Abbreviations are provided in the Abbreviation Listing.
a. Occasion 1 IOV Ka estimates shown. Occasion 2 shrinkage is 68.
b. Occasion 1 IOV relF estimates shown. Occasion 2 shrinkage is 56.6.
c. Values presented in parentheses are the final estimates converted to a %CV.

Figure 7: Standard goodness-of-fit plots for the final population PK model colored by indication



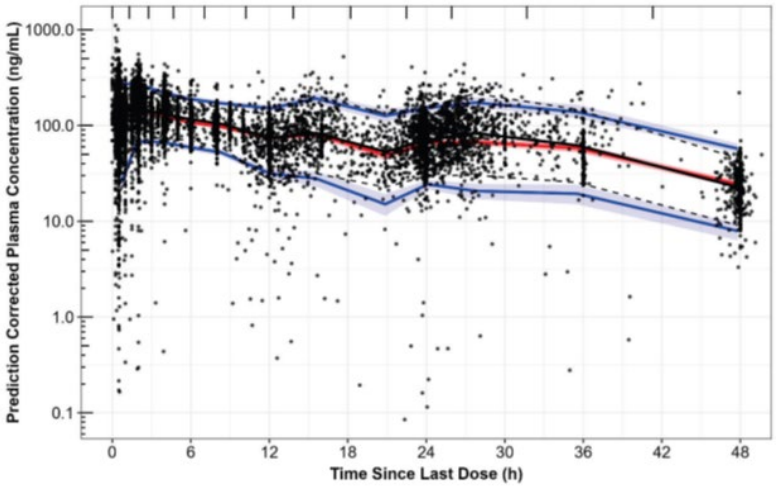
Note: Abbreviations are provided in the Abbreviation Listing. Note that concentrations (both observed and fitted) are presented on the natural log scale with units of ng/mL.

Figure 8: Prediction-corrected visual predictive check plot for the final population PK model using the pooled analysis dataset



Note: Abbreviations are provided in the Abbreviation Listing. Circles are observed concentrations, black solid lines are the median observed concentrations, black dashed lines are the 5th and 95th percentiles of the observed concentrations. Black tick marks at top of figure indicate the times since last dose at which the data were binned for the calculation of summary statistics. Red and blue shaded regions are the 90% confidence intervals for the median, 5th, and 95th percentiles from the simulations.

Figure 9: Prediction-corrected visual predictive check plot for the final population PK model using the pooled analysis dataset, truncated to the first 48 hours after a dose



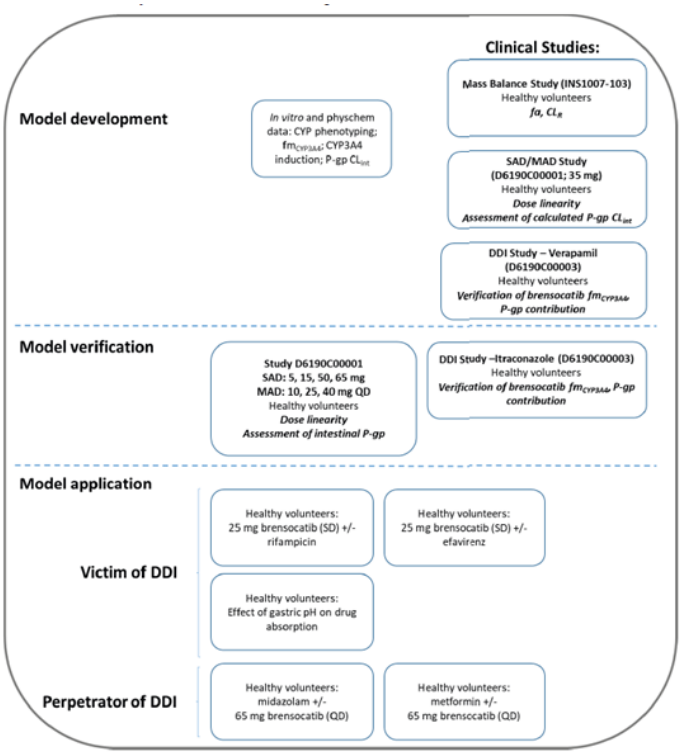
6.2.2.2.2. Physiology based pharmacokinetic model

The aim of this modelling was to develop a physiologically based pharmacokinetic (PBPK) model for brensocatib based on the available *in vitro* and clinical PK data to assess the DDI liability of brensocatib as a victim of CYP3A-mediated interactions in healthy subjects. Additionally, the impact of administration of brensocatib on the exposure (AUCinf and Cmax) of the CYP3A substrate midazolam and MATE substrate metformin in healthy subjects was evaluated.

As brensocatib displays pH-dependent solubility, the model was also prospectively applied to evaluate the effect of gastric pH on brensocatib disposition by simulating a single oral dose of brensocatib (40 mg) at elevated gastric pH (7.0) and default gastric pH (1.5).

The modelling for this project was split into three parts: model development, verification, and application.

Figure 10: Schematic illustrating the key PBPK modelling steps and components of each clinical study used in model building and verification



CL_{int}: intrinsic clearance; CL_R: renal clearance; fa: fraction absorbed; fm: fraction metabolised; SD: single dose.

Model development

A PBPK model that includes a mechanistic absorption model (Simcyp Advanced Dissolution, Absorption and Metabolism (ADAM) model) was developed. Input parameters from relevant *in-vitro* and *in-vivo* studies is shown in table below.

Table 12: PBPK Model Input Parameters for Brensocatib

PARAMETER	Value	Reference
Physicochemical and Binding Parameters		
MW (g/mol)	420.465	Data Checklist / IB v5, section 3.1
Log P	-2.1	Personal correspondence
Compound type	Base	SIVA
pKa	8.1	Data Checklist
B:P	0.7	INS1007-103 (draft report)
fu	0.128	ADME-AZS-Wave4-130621
Main binding protein	HSA	Assumed
Absorption Model –ADAM Model		
P_{app} ($x10^{-6}$ cm/s)	24.22	Optimised, SIVA
Calibrator P_{app} ($x10^{-6}$ cm/s)	Not provided	
P_{eff} ($x10^{-6}$cm/s)	2.64	Predicted, MechP_{eff}
$P_{trans,0}$ ($x10^{-6}$cm/s)	1527	Calibrated against Caco-2 P_{app} data
Q_{gut} (L/h)	11.76	Predicted
fu_{gut}	1.00	Assumed
ka (h^{-1})	1.15	Predicted
fa	0.976	Calculated, human mass balance data
F_G	1	Predicted from ADAM model
Regional Abs. Scalar – J1	2.5	Optimised to improve the prediction of C_{max}
Regional Abs. Scalar – J2	2.0	
Distribution Model – Minimal PBPK Model		

PARAMETER	Value	Reference
V_{SS} (L/kg)	1.24	Predicted, method 2
CL_{in} (L/h)	70.62	Optimised
CL_{out} (L/h)	51.93	Optimised
V_{SAC} (L/kg)	1.18	Optimised
Elimination Parameters		
CL/F (L/h)	5.595	PopPK estimate
$fm_{h,CYP3A4}$	0.20	Optimised value (Study D6190C00003)
$fm_{h,CYP2C8}$	0.036	ADME-AZS-Wave3-140531-CYP phenotyping
$fm_{h,CYP2D6}$	0.021	
CYP3A4 CL_{int} (µL/min/pmol)	0.013	Retrograde CL calculation
CYP2C8 CL_{int} (µL/min/pmol)	0.016	
CYP2D6 CL_{int} (µL/min/pmol)	0.022	
HLM CL_{int} (µL/min/mg)	5.983	
fu_{mic}	0.9812	Human hepatocyte metabolism BE000901_12
CL_R (L/h)	1.53	Study INS1007-103
Transport Parameters		
P-gp CL_{intT} (uL/min)	1.192	SIVA fitted value
RAF/REF	5	Optimised value (Study D6190C00003)
fu_{inc}	1	Default
Interaction Parameters		
MATE1 K_i (µM)	5.745	Report 16AZTrPISI
MATE-2K K_i (µM)	7.65	
fu_{inc}	1	Assumed
CYP3A4 Ind_{max}	1.75	Personal correspondence; draft data from Labcorp (Study 8492164)
CYP3A4 $IndC_{50}$ (µM)	3.24	

Model verification

Following satisfactory recovery of brensocatib plasma concentration-time profiles after single oral dose to healthy subjects, the final brensocatib PBPK model was applied to assess the recovery of observed PK profiles/exposures of brensocatib following single and repeat (for 28 days) oral doses of 5 – 65 mg QD.

Model simulations

Once the brensocatib PBPK model had been verified against the available clinical data, a series of DDI simulations were performed:

Victim of DDI:

- Simulation of plasma concentrations of brensocatib in healthy subjects following a single oral dose of 25 mg administered in the absence of rifampicin and on the 9th day of 16 days dosing with 600 mg rifampicin QD.
- Simulation of plasma concentrations of brensocatib in healthy subjects following a single oral dose of 25 mg administered in the absence of efavirenz and on the 9th day of 16 days dosing with 600 mg efavirenz QD.
- Simulation of plasma concentrations of brensocatib in healthy subjects following a single oral dose of 40 mg administered at elevated (pH 7.0) and default (pH 1.5) gastric pH.

Perpetrator of DDI:

- Prediction of plasma concentrations of midazolam in healthy subjects following a single oral dose of 5 mg administered in the absence of brensocatib and on the 8th day of 10 days of dosing with 65 mg brensocatib QD
- Prediction of plasma concentrations of metformin in healthy subjects following a single oral dose of 500 mg (390 mg free base equivalent dose) administered in the absence of brensocatib and on the 8th day of 10 days of dosing with 65 mg brensocatib QD.

Version 21 of the Simcyp Population-Based Simulator (www.simcyp.com) was used for all PBPK modelling and simulation.

Results

Model development

Simulation of a Single Oral Dose of 35 mg Brensocatib in Healthy Subjects (Base Model)

Table 13: Simulated and observed geometric mean PK parameters for brensocatib after a single 35 mg oral dose in healthy subjects.

	AUC_{inf} (nM*h)	C_{max} (nM)	t_{max}* (h)
Simulated	12773	637	1.05
CV%	39	22	0.70 – 2.45
Observed	13230	668.1	0.75
CV%	19.6	26.7	0.53 – 1.50
S/O	0.97	0.95	--

t_{max}: median, min, max; CV: Coefficient of Variation; S/O: Simulated/Observed; Source observed data: Study D6190C00001; Source simulated data: 07b-final-opt

Simulation of a Single Oral Dose of 25 mg Brensocatib Co-Administered with Itraconazole and Verapamil – Verification of P-gp Contribution and fmCYP3A4 (Final Model)

Table 14: Simulated and observed geometric mean AUCinf and Cmax values and corresponding GMRs for brensocatib in the absence and presence of verapamil in healthy subjects.

	Brensocatib		Brensocatib + Verapamil		GMR	
	AUC_{inf} (nM*h)	C_{max} (nM)	AUC_{inf} (nM*h)	C_{max} (nM)	AUC_{inf}	C_{max}
Simulated	8533	390	11066	550	1.30	1.41
CV%	39	31	35	20	--	--
90% CI	--	--	--	--	1.27 - 1.32	1.38 - 1.45
Observed	6697	385.8	8857	591.9	1.34	1.53
CV%	44.8	27.4	43.3	27.1	--	--
90% CI	--	--	--	--	1.23 – 1.45	1.37 – 1.73
S/O	1.27	1.01	1.25	0.93	0.97	0.92

CI: Confidence Interval; CV: Coefficient of Variation; GMR: geometric mean ratio; S/O: Simulated/Observed; Source observed data: Study D6190C00003; Source simulated data: 22c-verap-bren-opt-2.

Simulation of a Single Oral Dose of 35 mg Brensocatib in Healthy Subjects (Final Model)

Table 15: Simulated and observed geometric mean PK parameters for brensocatib after a single 35 mg oral dose in healthy subjects.

	AUC_{inf} (nM*h)	C_{max} (nM)	t_{max}* (h)
Simulated	12041	553	1.08
CV%	36	29	0.65 – 2.65
Observed	13230	668.1	0.75
CV%	19.6	26.7	0.53 – 1.50
S/O	0.91	0.83	--

t_{max}: median, min, max; CV: Coefficient of Variation; S/O: Simulated/Observed; Source observed data: Study D6190C00001; Source simulated data: v03b-35mg-sd

Model verification

Simulation of a Single Oral Doses of Brensocatib (5 – 65 mg) in Healthy Subject

Table 16: Simulated and observed geometric mean PK parameters for brensocatib after a single oral dose (5 – 65 mg) in healthy subjects.

	5 mg		15 mg		50 mg		65 mg	
	AUC_{inf} (nM*h)	C_{max} (nM)	AUC_{inf} (nM*h)	C_{max} (nM)	AUC_{inf} (nM*h)	C_{max} (nM)	AUC_{inf} (nM*h)	C_{max} (nM)
Simulated	1765	77.7	4936	232	16801	767	21413	987
CV%	36	28	37	28	39	31	37	30
Observed	915.3	50.74	4165	202.3	17590	1035	24710	1358
CV%	46.1	47.2	10.8	16	25.2	11.5	21.5	24.2
S/O	1.93	1.53	1.19	1.15	0.96	0.74	0.87	0.73

CV: Coefficient of Variation; S/O: Simulated/Observed; Source observed data: Study D6190C00001; Source simulated data: v01b-5mg-sd, v02b-15mg-sd, v04b-50mg-sd, v05b-65mg-sd.

Simulation of Multiple Oral Doses of Brensocatib (10 – 40 mg QD) in Healthy Subjects

Table 17: Simulated and observed geometric mean PK parameters for brensocatib after the first and multiple oral doses of 10 mg brensocatib QD for 21 days in healthy subjects.

	Day 1		Day 21	
	AUC_{0-24} (nM*h)	C_{max} (nM)	AUC_{last} (nM*h)	C_{max} (nM)
Simulated	2033	159	6043	254
CV%	25	29	55	29
Observed	1277	138.6	5135	246.3
CV%	18.9	18.3	35.1	26.2
S/O	1.59	1.15	1.18	1.03

CV: Coefficient of Variation; S/O: Simulated/Observed; Source observed data: Study D6190C00001; Source simulated data: v06b-10-mg-qd

Table 18: Simulated and observed geometric mean PK parameters for brensocatib after the first and multiple oral doses of 25 mg brensocatib QD for 28 days in healthy subjects.

	Day 1		Day 28	
	AUC_{0-24} (nM*h)	C_{max} (nM)	AUC_{last} (nM*h)	C_{max} (nM)
Simulated	4614	373	14355	602
CV%	25	27	58	29
Observed	3259	350.9	14320	598.1
CV%	15.9	19.3	44.2	15.5
S/O	1.42	1.06	1.00	1.01

CV: Coefficient of Variation; S/O: Simulated/Observed; Source observed data: Study D6190C00001; Source simulated data: v07b-25-mg-qd

Table 19: Simulated and observed geometric mean PK parameters for brensocatib after the first and multiple oral doses of 40 mg brensocatib QD for 28 days in healthy subjects.

	Day 1		Day 28	
	AUC_{0-24} (nM*h)	C_{max} (nM)	AUC_{last} (nM*h)	C_{max} (nM)
Simulated	7591	605	23301	968
CV%	29	31	55	31
Observed	6433	672	29550	1235
CV%	31	37	53.5	37.7
S/O	1.18	0.90	0.79	0.78

CV: Coefficient of Variation; S/O: Simulated/Observed; Source observed data: Study D6190C00001; Source simulated data: v08b-40-mg-qd

Simulation of a Single Oral Dose of 25 mg Brensocatib Co-Administered with Itraconazole – Verification of P-gp Contribution and fmCYP3A4 (Final Model)

Table 20: Simulated and observed geometric mean AUCinf and Cmax values and corresponding GMRs for brensocatib in the absence and presence of itraconazole in healthy subjects.

	Brensocatib		Brensocatib + Itraconazole		GMR	
	AUC_{inf} (nM*h)	C_{max} (nM)	AUC_{inf} (nM*h)	C_{max} (nM)	AUC_{inf}	C_{max}
Simulated	8691	391	11367	522	1.31	1.34
CV%	37	30	34	21	--	--
90% CI	--	--	--	--	1.29 - 1.33	1.31 - 1.36
Observed	6697	385.8	7615	234.1	1.12	0.61
CV%	44.8	27.4	35.1	39	--	--
90% CI	--	--	--	--	1.03 – 1.22	0.54 – 0.68
S/O	1.30	1.01	1.49	2.23	1.17	2.20

CI: Confidence Interval; CV: Coefficient of Variation; GMR: geometric mean ratio; S/O: Simulated/Observed; Source observed data: Study D6190C00003; Source simulated data: 23c-itra-bren-opt-2

Model Application – Victim DDI

Simulation of a Single Oral Dose of 25 mg Brensocatib Administered with Rifampicin in Healthy Subjects

Table 21: Simulated geometric mean AUCinf and Cmax values and corresponding GMRs for brensocatib in the absence and presence of rifampicin in healthy subjects

	Brensocatib		Brensocatib + Rifampicin		GMR	
	AUC_{inf} (nM*h)	C_{max} (nM)	AUC_{inf} (nM*h)	C_{max} (nM)	AUC_{inf}	C_{max}
Geometric Mean	8193	425	4779	393	0.583	0.926
90% CI – Lower	7749	405	4404	371	0.558	0.902
90% CI – Upper	8662	446	5186	417	0.609	0.950

GMR: geometric mean ratio; CI: Confidence Interval; Source simulated data: app-01b-rif-ddi

Table 22: Simulated geometric mean AUCinf and Cmax values and corresponding GMRs for brensocatib in the absence and presence of rifampicin in healthy subjects with induction of P-gp excluded.

	Brensocatib		Brensocatib + Rifampicin		GMR	
	AUC_{inf} (nM*h)	C_{max} (nM)	AUC_{inf} (nM*h)	C_{max} (nM)	AUC_{inf}	C_{max}
Geometric Mean	8466	427	5786	507	0.683	1.19
90% CI – Lower	8013	407	5429	487	0.662	1.16
90% CI – Upper	8945	448	6166	528	0.706	1.21

GMR: geometric mean ratio; CI: Confidence Interval; Source simulated data: app-08b-rif-ddi-nopgp

Simulation of a Single Oral Dose of 25 mg Brensocatib Administered with Efavirenz in Healthy Subjects

Table 23: Simulated geometric mean AUCinf and Cmax values and corresponding GMRs for brensocatib in the absence and presence of efavirenz in healthy subjects.

	Brensocatib		Brensocatib + Efavirenz		GMR	
	AUC_{inf} (nM*h)	C_{max} (nM)	AUC_{inf} (nM*h)	C_{max} (nM)	AUC_{inf}	C_{max}
Geometric Mean	8345	426	6457	409	0.774	0.960
90% CI – Lower	7889	406	6104	389	0.758	0.956
90% CI – Upper	8828	447	6830	429	0.790	0.963

GMR: geometric mean ratio; CI: Confidence Interval; Source simulated data: app-02b-efv-ddi

Simulation of a Single Oral Dose of 40 mg brensocatib administered at elevated (pH 7.0) and default (pH 1.5) gastric pH in healthy subjects

Table 24: Simulated geometric mean AUCinf and Cmax values for brensocatib at default (pH 1.5) and elevated (pH 7.0) gastric pH in healthy subjects

	AUC_{inf} (nM*h)	C_{max} (nM)
pH 1.5	13339	679
CV%	37	30
pH 7.0	13369	678
CV%	40	29
Ratio (pH 7 / pH 1.5)	1.00	1.00

CV: Coefficient of Variation. Source simulated data: app-03b-40mg-sd-ph1; app-04b-40mg-sd-ph7

Model Application – Perpetrator DDI

Simulation of a Single Oral Dose of 5 mg Midazolam Administered with Brensocatib in Healthy Subjects

Table 25: Simulated geometric mean AUCinf and Cmax values and corresponding GMRs for midazolam in the absence and presence of brensocatib in healthy subjects.

	Midazolam		Midazolam + Brensocatib		GMR	
	AUC_{inf} (nM*h)	C_{max} (nM)	AUC_{inf} (nM*h)	C_{max} (nM)	AUC_{inf}	C_{max}
Geometric Mean	170	59.4	161	56.8	0.949	0.956
90% CI – Lower	150	53.0	142	50.6	0.942	0.950
90% CI – Upper	193	66.6	183	63.7	0.955	0.962

GMR: geometric mean ratio; CI: Confidence Interval; Source simulated data: app-05c-mdz-ddi-65mg-qd

Simulation of a Single Oral Dose of 390 mg Metformin Administered with Brensocatib in Healthy Subjects

Table 26: Simulated geometric mean AUCinf and Cmax values and corresponding GMRs for metformin in the absence and presence of brensocatib in healthy subjects.

	Metformin		Metformin + Brensocatib		GMR	
	AUC_{inf} (nM*h)	C_{max} (nM)	AUC_{inf} (nM*h)	C_{max} (nM)	AUC_{inf}	C_{max}
Geometric Mean	49782	7072	50002	7089	1.00	1.00

GMR: geometric mean ratio; Source simulated data: app-06b-met-ddi-65mg-qd

6.2.2.3. Absorption

SAD / MAD Study D6190C00001

A phase I, randomised, single-blind, placebo-controlled, 2-part study to assess the safety, tolerability, pharmacokinetics, pharmacodynamics and food effect of single and multiple oral doses of INS1007 in healthy volunteers

SAD / MAD Study D6190C0001 was the First-in-human (FIH) study conducted with brensocatib, which was applied as an aqueous oral solution. The study was conducted in 2 parts (Part 1a = single ascending dose [SAD]; Part 1b = food effect and Part 2 = multiple ascending dose [MAD]). Escalation to the next higher dose only took place after review of the safety, tolerability, PK and PD (where applicable) data at each dose level by the safety Review Committee (SRC). Dose escalation was guided by specific stopping rules.

Up to 5 ascending dose levels were planned (with the possibility of adding 3 additional cohorts if needed). No additional cohorts were required. Within each of the cohorts 6 subjects were randomised to receive INS1007 and 3 subjects received placebo.

Part 1a (SAD)

Geometric mean plasma concentration-time profiles for INS1007 following single dose administration INS1007 at 5, 15, 35, 50 and 65 mg in Part 1a are shown in the Figure below.

Figure 11: Geometric Mean Plasma Concentrations (nmol/L) of INS1007 Versus Time Following Single INS1007 Administration at 5, 15, 35, 50 and 65 mg in Part 1a to Healthy Subjects under Fasted Conditions, Study D6190C00001

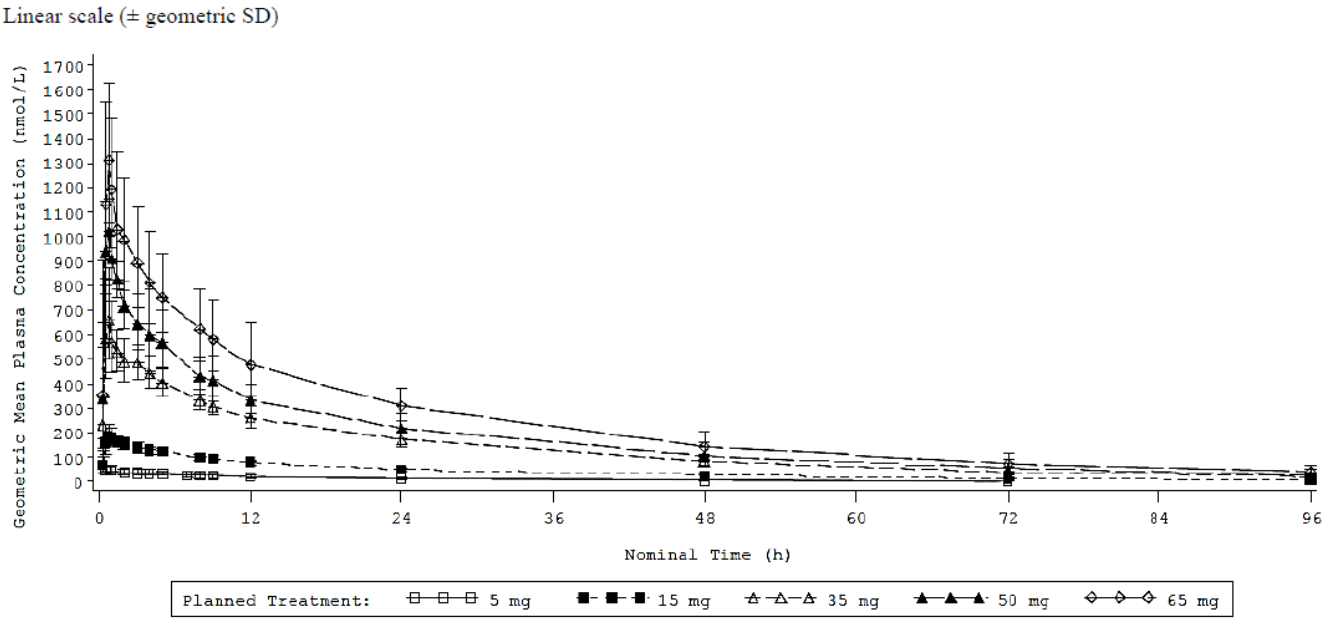


Table 27: Summary of INS1007 Pharmacokinetic Parameters by Treatment (Pharmacokinetic Analysis Set) Part 1a, Study D6190C00001

Parameter (Unit)		AZD7986 5 mg (n = 6)	AZD7986 15 mg (n = 6)	AZD7986 35 mg (n = 6)	AZD7986 50 mg (n = 6)	AZD7986 65 mg (n = 6)
$AUC_{(0\text{-last})}$ (h·nmol/L)	Geometric Mean	839.0*	3855	12430	16470	22880
	CV%	(48.3)	(9.3)	(18.3)	(22.7)	(21.2)
AUC (h·nmol/L)	Geometric Mean	915.3	4165	13230	17590	24710
	CV%	(46.1)	(10.8)	(19.6)	(25.2)	(21.5)
C_{max} (nmol/L)	Geometric Mean	50.74	202.3	668.1	1035	1358
	CV%	(47.2)	(16.0)	(26.7)	(11.5)	(24.2)
t_{max} (h)	Median	0.50	0.75	0.75	0.64	0.75
	Min, Max	(0.50, 1.00)	(0.53, 2.00)	(0.53, 1.50)	(0.52, 0.75)	(0.52, 1.98)
$t_{1/2\lambda z}$ (h)	Arithmetic Mean	19.96	25.92	23.80	24.52	25.50
	SD	(3.416)	(4.452)	(3.328)	(5.836)	(7.254)
MRT (h)	Arithmetic Mean	27.03	34.94	32.15	32.19	33.76
	SD	(4.373)	(5.863)	(5.003)	(8.360)	(10.35)
CL/F (L/h)	Arithmetic Mean	14.23	8.606	6.395	6.938	6.371
	SD	(7.577)	(0.9321)	(1.281)	(1.777)	(1.317)
Vz/F (L)	Arithmetic Mean	385.1	318.8	215.8	236.8	231.0
	SD	(129.6)	(46.72)	(29.62)	(39.64)	(67.56)

*t_{last} was nominal 48 hours for one subject, 72 hours for 4 subjects and 96 hours for one subject, therefore may not be comparable with 15 to 65 mg dose levels.

AUC: area under plasma concentration-time curve from zero extrapolated to infinity; $AUC_{(0\text{-last})}$: area under the plasma concentration-time curve from time zero to time of last quantifiable analyte concentration; CL/F: apparent clearance, C_{max}: observed maximum concentration; CV%: geometric coefficient of variation; max: maximum; min: minimum; n: number of subjects in the given category; SD: standard deviation; $t_{½\lambda z}$: half-life associated with terminal slope (λ_z) of a semi-logarithmic concentration-time curve; t_{max}: time to reach maximum observed concentration; λ_z: elimination rate constant, V_z/F: apparent volume of distribution.

Data Source: End-of-Text Table 14.2.4.

Following single dose administration of INS1007 at 5, 15, 35, 50 and 65 mg to healthy subjects under fasted conditions, the rate of INS1007 absorption was generally rapid with median tmax occurring at between 0.50 and 0.75 hours post-dose compared independent of dose level. After reaching Cmax, plasma INS1007 concentrations declined in a generally biphasic manner independent of dosage with a mean t½λz of 20.0, 25.9, 23.8, 24.5 and 25.5 hours following a 5, 15, 35, 50 and 65 mg dose, respectively.

Exposure (Cmax and AUC) increased in a supra-proportional manner between 5 mg and 35 mg doses but appear proportional over the 35 to 65 mg doses. Statistical analysis of the effect of single dose levels on the exposure, as measured by Cmax and AUC confirmed the trend for a slightly supra-proportional increase in exposure with slope estimates being 1.3, the increases were statistically significant as the lower and upper 95% CIs were 1.2 to 1.4, respectively.

Table 28: Power Model Dose-Proportionality Assessment of INS1007 Exposure versus Dose Following Single Dose Administration of INS1007 to Healthy Subjects under Fasted Conditions – Part 1a, Study D6190C00001

		Slope			Intercept			
Parameter (unit)	n	Estimate	SE	95% CI	Estimate	SE	95% CI	Coefficient of determination
C_{max} (nmol/L)	30	1.302	0.05056	(1.198, 1.406)	1.824	0.1681	(1.479, 2.168)	0.959
AUC (h*nmol/L)	30	1.287	0.05085	(1.183, 1.391)	4.798	0.1691	(4.452, 5.145)	0.958

CI: confidence interval; n: number of data points used in regression; SE: standard error.
Model: The linear model used was log (Y) = intercept + slope * log (dose), where Y is the parameter estimate; the natural logarithm has been used. Exponentiation of both sides of this equation produces the usual form of the power model: Y = exp(intercept) * (dose) %_slope.
The slope parameter is obtained via linear least squares.
Data Source: End-of-Text Table 14.2.16.

The fraction of the dose excreted in the urine as unchanged drug was moderate for all dose levels, with 11.1%, 13.5%, 19.9%, 16.5% and 18.0% being eliminated up to 48 hours post-dose at the 5, 15, 35, 50 and 65 mg single dose levels, respectively.

Part 2 (MAD)

In Part 2, participants received once daily oral administration of brensocatib at 10 mg, 25 mg or 40 mg or placebo for up to 28 days.

Table 29: Summary of INS1007 Pharmacokinetic Parameters by Treatment Following a Single INS1007 Dose on Day 1 and Daily Dosing on Days 21 or 28 at 10, 25 and 40 mg in Part 2 to Healthy Subjects under Fasted Conditions, Study D6190C00001

Parameter (Unit)		Day 1			Day 21	Day 28	
		AZD7986 10 mg (n = 6)	AZD7986 25 mg (n = 8)	AZD7986 40 mg (n = 10)	AZD7986 10 mg (n = 6)	AZD7986 25 mg (n = 7)	AZD7986 40 mg (n = 10)
$AUC_{(0\text{-last})}$ (h·nmol/L)	GeoMean	1278	3256	6427	5135	14320	29550
	CV%	(18.9)	(15.9)	(31.0)	49.4	39.5	53.5
AUC_{τ} (h·nmol/L)	GeoMean	1277	3259	6433	2834	7499	15660
	CV%	(18.9)	(15.9)	(31.0)	(35.1)	(27.4)	(43.3)
AUC (h·nmol/L)	GeoMean	1778[$]	4957[$]	9866[$]	5565	16660	33220
	CV%	(21.3)	(19.5)	(35.8)	54.3	44.2	58.5
C_{max} (nmol/L)	GeoMean	138.6	350.9	672.0	246.3	598.1	1235
	CV%	(18.3)	(19.3)	(37.0)	(26.2)	(15.5)	(37.7)
t_{max} (h)	Median	1.51	0.75	0.75	1.50	0.75	1.12
	Min, Max	(0.52, 1.53)	(0.53, 2.00)	(0.52, 1.50)	(0.75, 1.50)	(0.52, 2.00)	(0.53, 1.50)
$t_{1/2\lambda z}$ (h)	ArithMean	12.82[$]	14.85[$]	15.85[$]	25.85	33.78	30.04
	SD	(1.477)	(2.244)	(4.132)	(5.501)	(10.31)	(5.483)
MRT (h)	ArithMean	18.56	21.84	22.64	33.73	40.16	37.46
	SD	(2.245)	(3.067)	(5.844)	(9.663)	(10.58)	(8.157)
CL/F (L/h)	ArithMean	13.61	12.20	10.24	8.786	8.165	6.577
	SD	(2.665)	(2.485)	(4.206)	(2.772)	(2.025)	(2.867)
Vz/F (L)	ArithMean	248.9	256.7	222.7	311.8	393.2	269.6
	SD	(38.94)	(36.11)	(65.93)	(56.96)	(163.1)	(79.13)
Rac $_{AUC(0\text{-}\tau)}$	ArithMean	NA	NA	NA	2.257	2.330	2.476
	SD	NA	NA	NA	(0.4560)	(0.4828)	(0.4521)
Rac $_{Cmax}$	ArithMean	NA	NA	NA	1.799	1.667	1.881
	SD	NA	NA	NA	(0.3084)	(0.2007)	(0.4015)
TCP	ArithMean	NA	NA	NA	1.610	1.535	1.599
	SD	NA	NA	NA	(0.2539)	(0.2479)	(0.2082)

ArithMean: Arithmetic Mean; AUC: area under plasma concentration-time curve from zero extrapolated to infinity; AUC_{τ}: area under the plasma concentration-time curve from time zero to the end of the dosing interval (AUC from time zero to 24 hours post-dose presented on Day1); $AUC_{(0\text{-last})}$: area under the plasma concentration-time curve from time zero to time of last quantifiable analyte concentration; CL/F: apparent clearance, C_{max}: observed maximum concentration; CV%: geometric coefficient of variation; GeoMean: Geometric Mean; max: maximum; min: minimum; MRT: mean residence time; n: number of subjects in the given category; NA: not applicable; Rac $_{AUC(0\text{-}\tau)}$: Accumulation ratio for $AUC_{(0\text{-}\tau)}$; Rac $_{Cmax}$: Accumulation ratio for C_{max}; SD: standard deviation; TCP; temporal change parameter; $t_{½\lambda z}$: half-life associated with terminal slope (λ_z) of a semi-logarithmic concentration-time curve; t_{max}: time to reach maximum observed concentration; λ_z: elimination rate constant, V_z/F: apparent volume of distribution.

[$] Data calculated over a 24 hour period and is therefore not comparable with the Day 21 and Day 28 data. $AUC_{(0\text{-last})}$ and AUC not presented following repeated dosing due to accumulation which may mean the data is misleading.

Data Source: End-of-Text Table 14.2.6.1 and End-of-Text Table 14.2.6.2.

Exposure as measured by Cmax and AUCτ generally increased in a slightly supra-proportional manner between 10 mg and 40 mg daily doses, however in contrast to the single dose results dose-proportionality over the 10 to 25 mg doses was observed. Statistical analysis of the effect of single dose levels on the exposure, as measured by Cmax and AUC confirmed the trend for a slightly supra-proportional increase in exposure with slope estimates being 1.2, however the increases were not statistically significant as the lower and upper 95% CIs being 0.92 to 1.39 and 0.95 to 1.50, respectively.

Table 30: Power Model Dose-Proportionality Assessment of INS1007 Exposure versus Dose Following Multiple Daily Dose Administration of INS1007 to Healthy Subjects under Fasted Conditions – Part 2, Study D6190C00001

		Slope			Intercept			
Parameter (unit)	n	Estimate	SE	95% CI	Estimate	SE	95% CI	Coefficient of determination
C_{max} (nmol/L)	23	1.157	0.1098	(0.9285, 1.385)	2.794	0.3549	(2.056, 3.532)	0.841
AUC_{τ} (h*nmol/L)	23	1.228	0.1329	(0.9513, 1.504)	5.080	0.4297	(4.186, 5.973)	0.803

CI: confidence interval; n: number of data points used in regression; SE: standard error.
Model: The linear model used was log (Y) = intercept + slope * log (dose), where Y is the parameter estimate; the natural logarithm has been used.
Exponentiation of both sides of this equation produces the usual form of the power model: Y = exp(intercept) * (dose) %_slope.
The slope parameter is obtained via linear least squares.
Data Source: End-of-Text Table 14.2.17.

Following multiple once daily administration at 10 mg, 25 mg, and 40 mg, the steady state Cmax and AUCτ increased in an approximately dose proportional manner. There was 2.3- to 2.5- and 1.7- to 1.9-fold accumulation of INS1007 following multiple dosing, compared to the data from the single dose administration on Day 1 based on AUCτ and Cmax.

Single and Multiple dose Study incl. food effect – INS1007-101

A Phase I, Randomized, Double-Blind, Placebo-Controlled Study to Evaluate the Safety, Pharmacokinetics, and Pharmacodynamics of Single and Multiple Doses and the Open-Label Food Effect of a Single Dose of INS1007 in Healthy Japanese and Caucasian Subjects

The study was conducted sequentially in 2 parts.

Part A: Single and Multiple Doses

Up to 3 dose level cohorts (10, 25, and 40 mg) were planned to receive single and multiple oral doses of the IMP. Within each dose cohort, 10 Japanese and 10 Caucasian subjects were randomized in a 4:1 ratio to receive the IMP (i.e., 8 on INS1007 and 2 on placebo in each Japanese/Caucasian group).

Subjects received INS1007 or matching placebo administered daily from Day 4 to Day 30. Dosing occurred after overnight fasting of at least 10 hours. Day 29 and Day 30 were an in-house stay at the Clinical Unit.

On Day 1, after overnight fasting for at least 10 hours, a single dose of INS1007 or matching placebo was administered orally. Cohorts 1 (10 mg) and 2 (25 mg) were conducted in parallel. Escalation to the 40 mg dose (Cohort 3) did not begin until the available safety and tolerability data, through Day 33, of at least 6 Japanese and 6 Caucasian subjects in the 25 mg dose cohort were evaluated and deemed acceptable by both the Investigator and the Sponsor's Medical Monitor.

Doses were administered orally to subjects. The 40 mg dose was achieved by administering 15 mg and 25 mg tablets together.

For Part A, the INS1007 absorption was relatively rapid with plasma concentrations observed at the first time points taken post-dose (0.5 hours). Median Tmax occurred between 1 to 2 hours after single and repeat doses across dose levels and Japanese/Caucasian groups. The 10, 25, and 40 mg doses had similar elimination phases for both Japanese and Caucasian subjects.

Following single dose administration of 10, 25, and 40 mg INS1007, the PK parameters for Japanese subjects were compared to those for the Caucasian subjects. The 10 and 25 mg results indicate that the geometric mean Cmax, AUC0-τ, AUC0-last, and AUC0-inf were in a similar range, but the 40 mg had higher results for Caucasian subjects as compared to Japanese subjects on Day 1 (single dose).

Table 31: Summary of Pharmacokinetic Parameters for INS1007 Following Single Oral Dose Administration (10, 25, and 40 mg) to Japanese and Caucasian Subjects

Parameter (Unit)	Statistic	10 mg		25 mg		40 mg	
		Japanese	Caucasian	Japanese	Caucasian	Japanese	Caucasian
C_{max} (ng/mL)	n	8	8	9	8	8	8
	GeoMean	63.65	54.01	199.1	150.4	234.6	304.6
	CV% GeoMean	17.6	29	35.8	41.8	32.6	45.3
T_{max} (h)	n	8	8	9	8	8	8
	Median	1.02	1.01	1.50	1.35	1.50	1.27
	Range	0.75 - 2.00	1.00 - 2.00	0.75 - 8.00	0.50 - 3.00	0.77 - 3.02	0.50 - 7.78
$AUC_{0-\tau}$ (h*ng/mL)	n	8	8	9	8	8	8
	GeoMean	598.6	610.1	1942	1686	2534	3350
	CV% GeoMean	33.4	19.7	28.0	21.1	33.5	23.0
AUC_{0-last} (h*ng/mL)	n	8	8	9	8	8	8
	GeoMean	871.4	974.6	2899	2763	3804	5247
	CV% GeoMean	44.0	18.0	33.8	19.7	38.2	15.4
AUC_{0-inf} (h*ng/mL)	n	8	8	9	8	8	8
	GeoMean	935.9	1118	3194	3155	4228	5888
	CV% GeoMean	48.0	19.1	38.4	19.9	44.3	12.4
$AUC_{\%extrap}$ (%)	n	8	8	9	8	8	8
	GeoMean	5.818	10.56	8.156	11.68	8.525	9.230
	CV% GeoMean	70.8	69.1	57.5	33.7	72.6	63.5
λ_z (1/h)	n	8	8	9	8	8	8
	GeoMean	0.03764	0.02831	0.03246	0.02857	0.03093	0.03221
	CV% GeoMean	21.7	33.1	20.7	15.3	29.7	25.1
$t_{½}$ (h)	n	8	8	9	8	8	8
	GeoMean	18.42	24.49	21.35	24.26	22.41	21.52
	CV% GeoMean	21.7	33.1	20.7	15.3	29.7	25.1

CV% = coefficient of variation; GeoMean = geometric mean; n = number of subjects in the specific category
Data source: Table 14.2.2.1

Statistical Evaluation of Dose Proportionality

For the Caucasian subjects, the AUC parameters increase with dose was more than dose proportional on Day 1; by Day 30 the AUC increases are dose proportional. Cmax increased in a dose proportional manner on both Day 1 and Day 30 for Caucasian subjects. For the Japanese subjects, the AUC and Cmax parameters increased in a dose proportional manner on Day 1 and Day 30.

Table 32: Statistical Analysis of Dose Proportionality by Japanese/Caucasian Group – Day 1 and Day 30, Study INS1007-101

Descent/ Day	Parameter (Unit)	N	Slope	90% Confidence Interval
Caucasian Day 1	C_{max} (ng/mL)	24	1.23	(1.00, 1.46)
	$AUC_{0-\tau}$ (h*ng/mL)	24	1.21	(1.08, 1.34)
	AUC_{0-last} (h*ng/mL)	24	1.20	(1.10, 1.31)
	AUC_{0-inf} (h*ng/mL)	24	1.19	(1.08, 1.29)
Japanese Day 1	C_{max} (ng/mL)	25	0.99	(0.79, 1.18)
	$AUC_{0-\tau}$ (h*ng/mL)	25	1.08	(0.88, 1.27)
	AUC_{0-last} (h*ng/mL)	25	1.10	(0.87, 1.33)
	AUC_{0-inf} (h*ng/mL)	25	1.13	(0.87, 1.38)
Caucasian Day 30	C_{max} (ng/mL)	24	1.17	(1.00, 1.34)
	$AUC_{0-\tau}$ (h*ng/mL)	24	1.20	(1.00, 1.40)
	AUC_{0-last} (h*ng/mL)	24	1.15	(0.93, 1.36)
	AUC_{0-inf} (h*ng/mL)	24	1.12	(0.88, 1.36)
Japanese Day 30	C_{max} (ng/mL)	23	1.08	(0.87, 1.30)
	$AUC_{0-\tau}$ (h*ng/mL)	23	1.05	(0.77, 1.33)
	AUC_{0-last} (h*ng/mL)	23	1.05	(0.72, 1.37)
	AUC_{0-inf} (h*ng/mL)	23	1.04	(0.69, 1.39)

N = number of subjects

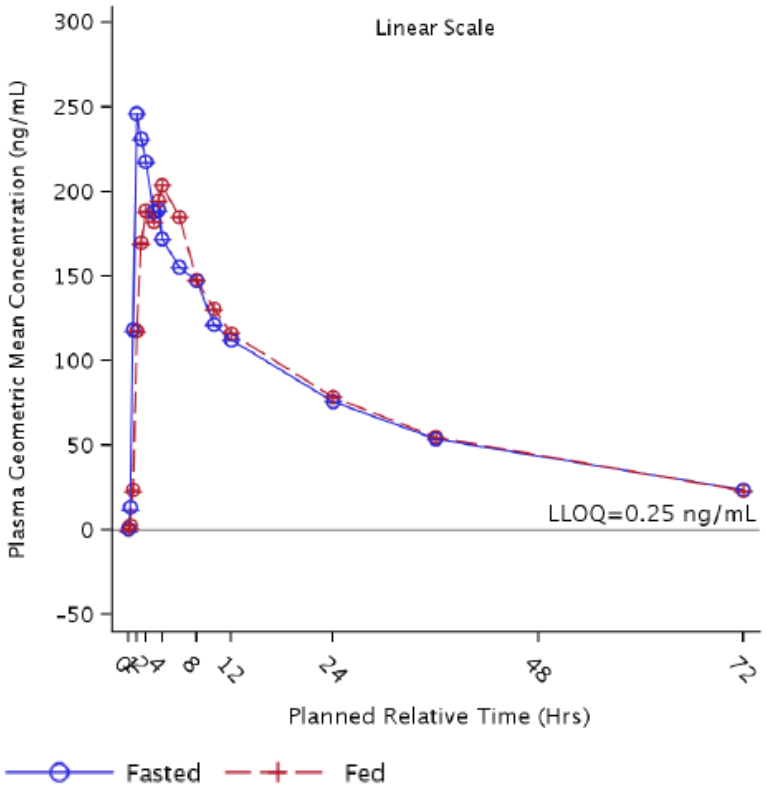
Dose proportionality was assessed using POWER model on single and multiple dose pharmacokinetic parameters independently.

Data source: Table 14.2.4.1

Part B: Food Effect

Part B evaluated the effect of a high-fat meal on the safety, tolerability, and PK of an oral dose of INS1007 40 mg. For this study part, 20 never-before-enrolled subjects (10 Japanese and 10 Caucasian) were randomized to one of the 2 treatment sequences in a 1:1 ratio. A washout period of 7 days between Period 1 and Period 2 (from Day 1 to Day 7).

Figure 12: Mean Plasma INS1007 Concentration Profiles in Caucasians After Administration of a 40 mg Dose in Fed and Fasted States (Linear Scale)



Comparing single administration of 40 mg INS1007 to Caucasian subjects in the fed versus fasted state, the presence of food caused a slightly delayed absorption plasma profile. The INS1007 median Tmax shifted from 1.25 to 2.00 hours fasted to fed, respectively.

In Caucasian subjects, the fed/fasted GMR demonstrated that the Cmax in the fed state was similar to the fasted state, but the 90% CI was slightly above the upper boundary of 125. The fed/fasted GMR for AUC0-last, and AUC0-inf demonstrated that the AUC in the fed state was equivalent compared to the fasted state. These data indicate a negligible food effect in Caucasian subjects.

Table 33: Statistical Analysis of Food Effect by Japanese/Caucasian Group, Study INS1007-101

Descent/ Number of Subjects	Parameter (Unit)	LS-mean		Geometric Mean Ratio (%)	90% Confidence Interval
		INS1007 40 mg Fed	INS1007 40 mg Fasted		
Caucasian (N=10)	C_{max} (ng/mL)	272.20	268.86	101.24	(81.41, 125.91)
	$AUC_{0\text{-last}}$ (ng*hr/mL)	5214.17	5146.58	101.31	(96.80, 106.04)
	$AUC_{0\text{-inf}}$ (ng*hr/mL)	6177.69	6201.24	99.62	(94.60, 104.90)
Japanese (N=9)	C_{max} (ng/mL)	292.99	355.80	82.35	(67.86, 99.93)
	$AUC_{0\text{-las)}}$ (ng*hr/mL)	4840.23	4918.93	98.40	(94.43, 102.53)
	$AUC_{0\text{-inf}}$ (ng*hr/mL)	5232.03	5273.96	99.21	(95.71, 102.83)

N = number of subjects exposed to each treatment who were included in the mixed model

Test: INS1007 40 mg under fed conditions; Reference: INS1007 40 mg under fasted conditions

Food effect was analyzed using analysis of variance with sequence, treatment group and period as fixed effects and subject (sequence) as random effect, after logarithmic transformation of the data.

Data source: Table 14.2.3.1

Solubility of brensocatib

A saturated solubility study was conducted using aqueous solutions with different pHs at 37±1ºC as shown in the Table below. Three replicate determinations for each test solution condition were measured, and the pH saturated solution was measured at the end of the equilibrium solubility study (24 hrs). The total degradation product of the solution was also determined.

In Vitro Permeability Studies

Caco-2 cell permeability Study BJAA-0002-DV-PB

For INS1007 at 10 µM, the apparent permeability (Papp) values in the Caco-2 cell assay were 9.03 x 10^{-6} cm/s in the apical-to-basolateral (A-B) direction, and 34.9 x 10^{-6} cm/s in the basolateral-to-apical (B-A) direction after a 2-hour incubation. The Papp A-B value places INS1007 in the higher permeable binning category.

The efflux ratio (ratio of the Papp value in the B-A direction divided by the A-B direction) was 3.86, indicating efflux by the Caco-2 cells for 10 µM INS1007 (the criterion for efflux used in this study was >2).

6.2.2.4. Bioequivalence

Formulations used during clinical development

Three different formulations were applied during clinical development, comprising an oral solution, used during early SAD/MAD Study D6190C00001 and DDI Study D6190C00003, a phase 2 and phase 3 film-coated tablet formulation.

The intended commercial formulation of 25 mg is identical for the core tablet formulation, shape, and size used across the clinical development program, except for changes in the film coating colour from brown used in the clinical studies to grey for the commercial drug product.

Dissolution studies were conducted to compare the dissolution profiles of Phase 2 with Phase 3 and Phase 3 with commercial drug product at pHs of 1.2, 4.5, and 6.8. The formulation comparability for the Phase 2 tablets, Phase 3 tablets, and the intended commercial formulation has been demonstrated by calculating the similarity factor f2 of the dissolution profiles or by visual comparison. The similarity factor f2 was calculated using the formula and following the conditions for the evaluation of the similarity factor as provided in International Council for Harmonisation M9.

Dissolution Comparison for 25 mg Tablets

The dissolution method used in the comparison of the dissolution profiles (n=12) of 25 mg tablet had 500 mL dissolution media at pH 1.2, pH 4.5, and pH 6.8 and the following apparatus:

- USP Apparatus II (paddle) at 50 rpm
- USP Apparatus II (paddle) at 65 rpm
- USP Apparatus I (basket) at 100 rpm

In conclusion, the changes between Phase 3 and commercial tablets were assessed for 25 mg tablets, and the similarity of the dissolution profiles was demonstrated by the results of f1 and f2, which shows that the Phase 3 and the proposed commercial tablets are comparable.

6.2.2.5. Distribution

In vitro data showed that brensocatib is moderately bound to human plasma proteins (87.2% bound) and there was negligible covalent binding to proteins following incubation in human hepatocytes.

In Study INS1007-105, the f_{unb} brensocatib was 16.9%, 19.7%, and 26.9% in participants with mild (Child-Pugh score 5 to 6), moderate (Child-Pugh score 7 to 9), and severe (Child-Pugh 10 to 12) HI, compared to 17.8% (ie, 82.2% bound) in healthy control participants. The f_{unb} was found to be highly correlated to the serum albumin levels in the participants, suggesting that brensocatib is mainly bound to albumin.

Based on the total radioactivity data from Study INS1007-103, the whole B/P of 14C-brensocatib was 0.716, suggesting insignificant binding to blood cells.

In healthy volunteers, mean apparent volume of distribution at terminal phase after oral administration (Vd/F) ranged from 216 to 413 L, similar to that in NCFBE and CF participants. In participants with severe RI or severe HI, the Vd/F appeared to be higher. The Vd/F data indicate that brensocatib is extensively distributed in the body.

Table 34: Summary of Mean Volume of Distribution in Participants from Clinical Studies

Study	Dose (mg) (n)	Single Dose V_d/F (L)	Steady-State V_d/F (L)
D6190C00001	Single dose (Part 1): 5, 15, 35, 50, 65 (6/dose) QD (Part 2): 10 (6), 25 (8), 40 (10)	385, 319, 216, 237, 231	312, 393[a], 270
D6190C00003	25 (15, brensocatib alone)	297	NA
INS1007-101, Caucasian	Part A: 10, 25, 40 (8/dose)	ND	349, 315, 228
INS1007-101, Japanese	Part A: 10 (8), 25 (9 on Day 1 and 8 on Day 30), 40 (8 on Day 1 and 7 on Day 30)	ND	329, 223, 355
INS1007-102	25 in mild, moderate, and severe RI (6/group) and healthy control (10)	397, 359, 492, 411	NA
INS1007-104	Part 1: 80 (6), 120 (6) Part 2: 40 (26), 120 (32)	Part 1: 243, 231; Part 2: 301, 267	NA
INS1007-105	25 in mild, moderate, and severe HI (6/group) and healthy control (9)	353, 396, 515, 413	NA
INS1007-106	25 (16 and 16 for Part 1 and Part 2, brensocatib alone)	314, 296	NA
INS1007-109	25 (22, brensocatib alone)	370	NA
INS1007-201[b]	QD: 10 (75), 25 (82)	NA	138, 138
INS1007-211	Day 1: 10, 25 and 40 (8/dose); Day 28 10 (8), 25 (7) and 40(8)	NA	423, 332, 398[a]
INS1007-301, adult[b]	QD: 10 (288), 25 (307)	NA	126, 131
INS1007-301, adolescent[b]	QD: 10 (15), 25 (16)	NA	83.6, 71.3

Source: Section 2 Table 2, Table 3, Table 4, Table 7, Table 8, Table 10, Table 12, Table 14, Table 21, and Table 24

[a] n = 7.

[b] Population PK model predicted (ICPD 00694-1).

6.2.2.6. Metabolism

Mass Balance Study INS1007-103

A Phase 1, Open-label Study of the Absorption, Metabolism, and Excretion of [14C]-Brensocatib Following a Single Oral Administration in Healthy Male Subjects

Seven participants received a single oral dose of 14C-brensocatib at 40 mg (approximately 100 µCi). Blood, plasma, urine, and feces samples were collected at selected timepoints or intervals for 13 days. The PK parameters and mass balance were determined based on individual data while metabolite profiling was based on the data from pooled samples.

Following administration of a single oral dose of 40 mg brensocatib containing approximately 100 µCi of [14C]-brensocatib, brensocatib was characterized by rapid absorption into the systemic circulation, with a median tmax value of 1.00 hour post-dose in plasma (range: 0.500 to 1.00 hour post-dose). After reaching Cmax, plasma concentrations appeared to decline in a biphasic manner, with an arithmetic mean t1/2 of 28.6 hours (range: 18.9 to 38.2 hours). Plasma concentrations of brensocatib were quantifiable for all 7 subjects at 120 hours post-dose and in 2 of 7 subjects at 240 hours post-dose.

Table 35: Summary of the PK Parameters for Brensocatib in Plasma and Total Radioactivity in Plasma and Whole Blood, Pharmacokinetic Population (Study INS1007-103)

PK Parameter	Unchanged Brensocatib	Total Radioactivity	
	Plasma	Plasma	Whole Blood
AUC_{last} (h*ng/mL)[a]	5410 (29.3)	41900 (20.2)	30100 (23.7)
AUC_{∞} (h*ng/mL)[a]	5460 (29.4)	NC	NC
C_{max} (ng/mL)[a]	259 (27.3)	384 (29.0)	238 (29.5)
T_{max} (h)	1.00 (0.500-1.00)	2.00 (1.00-6.03)	1.00 (1.00-6.03)
$t_{½}$ (h)	28.6 (26.9)	164 (19.7)	177 (16.5)

Source: Study INS1007-103 [Table 14.2.1.2]
[a] Units for total radioactivity AUCs and C_{max} are (h*ng-eq/mL) and (ng-eq/mL), respectively.
Note: Mean (CV%) presented; for T_{max}, median (min-max) presented.

The mean amounts of radioactivity recovered in urine and feces over 13 days post-dose were 54.2% (range: 51.6% to 59.3%) and 28.3% (range: 23.3% to 34.5%) of dose, respectively, and in total 82.5% (range: 80.0% to 86.3%). Most of the radioactivity was excreted within the first 72 hours (56.7%).

Unchanged brensocatib was a major component in plasma (16.2%) and urine (22.8%), but minor component in feces (2.41%).

Table 36: Summary of the Recovery of Total Radioactivity in Urine, Feces, and Overall Within 312 Hours Following a Single Oral Administration of 40 mg (100 µCi) [14C]-Brensocatib

	0 to 312 Hours Postdose
	Cumulative Amount (%)
Analyte (Matrix)	[Mean (range)]
Total Radioactivity (Urine)	54.2 (51.6-59.3)
Total Radioactivity (Feces)	28.3 (23.3-34.5)
Total Radioactivity (Overall)	82.5 (80.0-86.3)

Source: Labcorp Early Development Laboratories Inc. Study 8462138.
Protocol Reference: INS1007-103.

Brensocatib underwent moderate metabolism, primarily via oxidation, hydrolysis, oxidative dealkylation, sulfuration, and carbamoyl glucuronidation. A total of 27 quantifiable metabolites were detected in plasma, urine, and feces, and the chemical structures of 9 of them were identified. In plasma, 6 metabolites were detected, including M8 (thiocyanate), M41 (des(oxazepane carbaldehyde)-dehydro-brensocatib), M20 (desoxazepane-N-formylbrensocatib-), M59 (brensocatib carbamoyl glucuronide), M33 (oxydehydro-brensocatib), and M17 (dioxy-brensocatib-).

Among them, the M8 thiocyanate was the only major metabolite detected (51% of total plasma radioactivity, based on AUClast), and all other metabolites were minor (each <0.5% of total AUClast). Unchanged brensocatib accounted for 16.2% of total AUClast. The high contribution of the M8 metabolite to total plasma radioactivity AUC is understood to be mainly due to M8's extremely long half-life (7-8 days). All other metabolites identified were minor (individual AUC < 1% of total plasma radioactivity AUC) and were unquantifiable at 24 hours post-dose as well as at later time points. Besides M8, no other late forming metabolites were detected in plasma.

The metabolic profile of brensocatib in plasma, urine, and feces was characterized over 13 days (312 hours sampling). Like in plasma, in urine and feces, there were no late forming metabolites detected based on the data from pooled urine samples (0-240, 48-96, 120-168 and 192-312 hours) and feces samples (0-216, 48-96, 120-168 and 192-312 hours), and all quantifiable metabolites were present at 48-96 hour in urine and/or feces samples.

M8 was first detected in a 24 hour post-dose (also Tmax), and the overall amount of M8 formed was estimated to be <5% (0.11% in early pooled urine sampling [0-240 h], 0.806% in last sample [192-312 h], plus 3.49% estimated unrecovered [<312 h]).

Table 37: Radioactive components present in the last pooled excreta samples (192-312 hours) and extrapolated total amount formed

Matrix	ID	%·Sample·Activity·(192-312·h)	%·of·Radioactive·Dose			
			Last·sample·[a]·(192-312·h)	Pooled·Sample·(0–240·h·or·0-216·h)	Estimated·[b]·Unrecovered·(>312·h)	Estimated·Total·(0–>312·h)
Urine	Brensocatib	7.5	0.207	22.8	0.90	23.90
	M8	29.2	0.806	0.11	3.49	4.41
	M13	21.7	0.599	2.32	2.59	5.51
	M39	36.7	1.01	3.08	4.38	8.47
Faeces	Brensocatib	7.32	0.0653	2.41	0.55	3.02
	M13	27.6	0.247	1.26	2.06	3.57
	M39	30.1	0.268	0.999	2.25	3.52
	M49	13.0	0.116	0.630	0.97	1.72

Source: Report 8462137 Table 8, Table 9, Table 10, Table 11, Table 12, Table 14.
[a] Last sample radioactivity: 3.15% of dose in urine and 1.70% of dose in feces; total 4.85% of dose (urine = 3.15%/4.85% = 65% and feces = 1.70%/4.85% = 35%)
[b] Estimated unrecovered amount in urine (% of dose) = 17.5% x % sample activity (192-312h) x 65% x CF; Estimated unrecovered amount in feces (% of dose) = 17.5% x % sample activity (192-312h) x 35% x CF; CF is the correction factor (1.05 for urine and 1.22 for feces) to account for the unquantified sample radioactivity (4.9% in the urine sample and 12% in the feces sample).
CF = correction factor; ID = identifier; M8 = thiocyanate; M13 = descyano-brensocatib carboxylic acid; M39 = des(oxazepane carbaldehyde)brensocatib-carboxylic acid; M49: structure unknown

The effect of brensocatib on thiocyanate exposure was evaluated using selected PK samples from Studies INS1007-102 (25 mg in RI and HP), QT-Study INS1007-104 (120 mg or placebo in HP), INS1007-105 (25 mg in HI and HP), and INS1007-201 (25 mg or placebo in NCFBE participants).

Summary of Thiocyanate Evaluation Studies

In the mass balance and metabolic profiling study (Study INS1007-103), a major circulating metabolite, M8 (thiocyanate), was detected in HP following a single oral dose of ^{14}C-brensocatib at 40 mg (100 µCi).

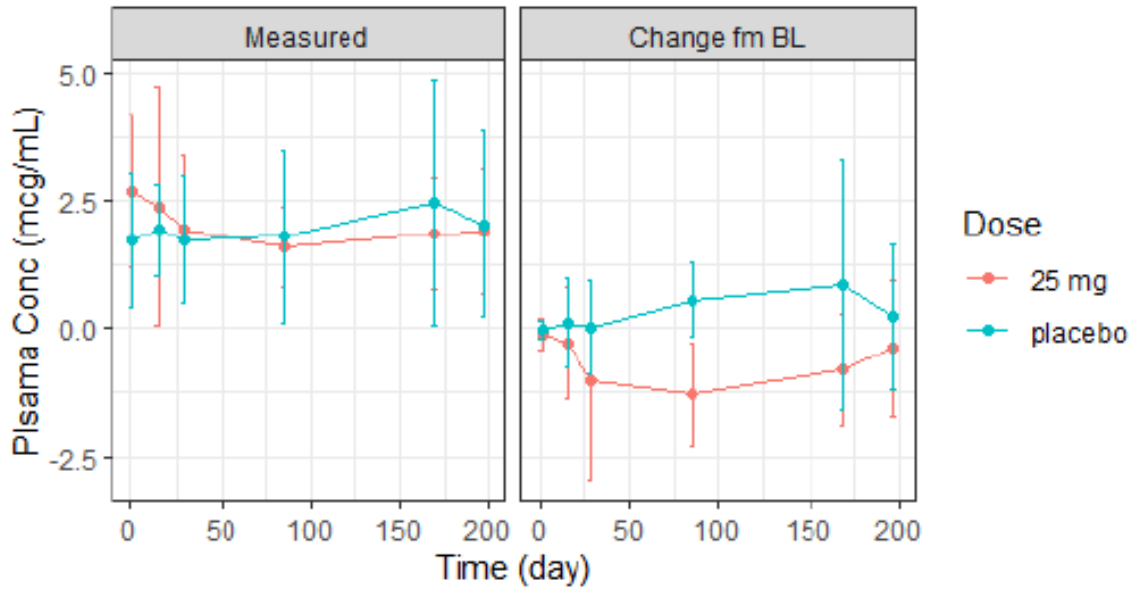
Thiocyanate is an endogenous and a *Generally recognized as safe* GRAS compound (GRAS Notification Sodium Thiocyanate, 2017) with a normal range of thiocyanate of 3 to 15 µg/mL in serum (prescribing information for Sodium Nitroprusside).

Selected PK samples were taken from NCFBE patients participating in phase study 201. Following QD administration of 25 mg brensocatib or placebo, the plasma concentrations of thiocyanate remained relatively consistent over the 24-week treatment period in NCFBE participants. The average thiocyanate concentrations in plasma were comparable in participants with QD brensocatib 25 mg or placebo. The highest thiocyanate plasma concentrations in brensocatib- and placebo-treated participants were 6.52 and 7.90 µg/mL, respectively, none of them above the upper limit of the normal range (ie, 15 µg/mL).

Thiocyanate serum concentrations are known to be affected by diet and smoking (GRAS Notification Sodium Thiocyanate, 2017).

Baseline-corrected concentrations were generally higher in participants receiving placebo than in treated participants and did not show any time-dependent elevation. The results indicate that brensocatib long term treatment does not have any noticeable effect on thiocyanate exposure in NCFBE participants and the observed concentration levels are primarily endogenous in nature.

Figure 13: Mean (± SD) Plasma Concentration Profiles of Thiocyanate in Participants With NCFBE Following QD Administration of Brensocatib 25 mg or Placebo for 24 Weeks



6.2.2.7. Elimination

Following a single dose, the mean values of total clearance after oral administration (CL/F) of brensocatib in healthy volunteers were generally <11 L/h (range: 6.4 to 10.7 L/h), except at 5 mg (14.2 L/h). The elimination t½ in HP after a single dose was approximately 20 to 30 hours (range: 18.8 to 30.3 hours), except for the healthy control participants in Study INS1007-102 (39.1 hours).

Steady-state CL/F and elimination t½ in healthy participants (HP) were comparable to those after a single dose, indicating the elimination of brensocatib is time-invariant, which is supported by the results of the population PK analyses. Compared to HP, NCFBE patients appeared to have lower CL/F and longer t½, while the CL/F and t½ in CF participants were generally comparable to those in HP.

Urinary excretion was evaluated in 3 studies, Studies D6190C00001, INS1007-103, and INS1007-105. Following a single dose of 5 to 65 mg, 11% to 20% of the brensocatib dose was excreted in urine as unchanged drug. With repeated QD dosing (10 to 40 mg), urinary excretion was 20.1% to 26.3% within 24 hours and 31.4% to 38.0% within 48 hours. Renal clearance was 1.43 to 1.73 L/h following a single dose and 1.51 to 1.62 L/h at steady-state, approximately 20% of CL/F.

Following a radiolabeled dose at 40 mg, 54.2% of total radioactivity was excreted in urine and 28.3% of total radioactivity was excreted in feces. Unchanged brensocatib excreted in urine and feces was 22.8% and 2.41% of dose, respectively.

Table 38: Mean CL/F and Elimination t½ From Clinical Studies

Study	Dose (mg) (n)	Single Dose		Steady-State	
		CL/F (L/h)	t½ (h)	CL/F (L/h)	t½ (h)
D6190C00001	Single dose (Part 1): 5, 15, 35, 50, 65 (6/dose) QD (Part 2): 10 (6), 25 (8 on Day 1, 7 on Day 28), 40 (10)	14.2, 8.6, 6.4, 6.9, 6.4	20.0, 25.9, 23.8, 24.5, 25.5	8.79, 8.17, 6.58	25.9, 33.8, 30.0
D6190C00003	25 (15, brensocatib alone)	9.81	23.4	NA	NA
INS1007-101, Caucasian	Part A: 10, 25, 40 (8/dose)	ND	25.7, 24.5, 22.1	8.62, 7.63, 6.80	28.1, 28.7, 23.6
INS1007-101, Japanese	Part A: 10 (8), 25 (9 on Day 1 and 8 on Day 30), 40 (8 on Day 1 and 7 on Day 30)	ND	18.8, 21.7, 23.2	10.8, 6.89, 11.4	22.5, 23.3, 21.9
INS1007-102	25 in mild, moderate, and severe RI (6/group) and healthy control (10)	7.72, 6.09, 9.97, 7.43	37.0, 42.8, 36.3, 39.1	NA	NA
INS1007-104	Part 1: 80 (6), 120 (6) Part 2: 40 (26), 120 (32)	Part 1: 7.02, 7.15 Part 2: 9.31, 8.11	Part 1: 26.2, 23.6 Part 2: 24.3, 25.2	NA	NA
INS1007-105	25 in mild, moderate, and severe HI (6/group) and healthy control (9)	7.58, 9.83, 13.3, 10.7	31.4, 27.9, 28.5, 30.3	NA	NA
INS1007-106	25 (16 and 16 for Part 1 and Part 2, brensocatib alone)	8.24, 8.74	27.2, 25.3	NA	NA
INS1007-109	25 (22, brensocatib alone)	11.4	25.0	NA	NA
INS1007-201[a]	QD: 10 (75), 25 (82)	NA	NA	2.58, 2.88	39.6, 35.7
INS1007-211	Day 1: 10, 25 and 40 (8/dose); Day 28: 10 (8), 25 (7) and 40 (7)	NA	NA	9.91, 7.17, 7.20	30.0, 33.1, 38.5
INS1007-301, adult[a]	QD: 10 (288), 25 (307)	NA	NA	2.81, 3.02	33.7, 32.6
INS1007-301, adolescent[a]	QD: 10 (15), 25 (16)	NA	NA	2.36, 2.07	27.8, 26.9

Source: Section 2 Table 2, Table 3, Table 4, Table 7, Table 8, Table 10, Table 12, Table 14, Table 21, and Table 24

[a] Population PK model predicted (ICPD 00694-1).

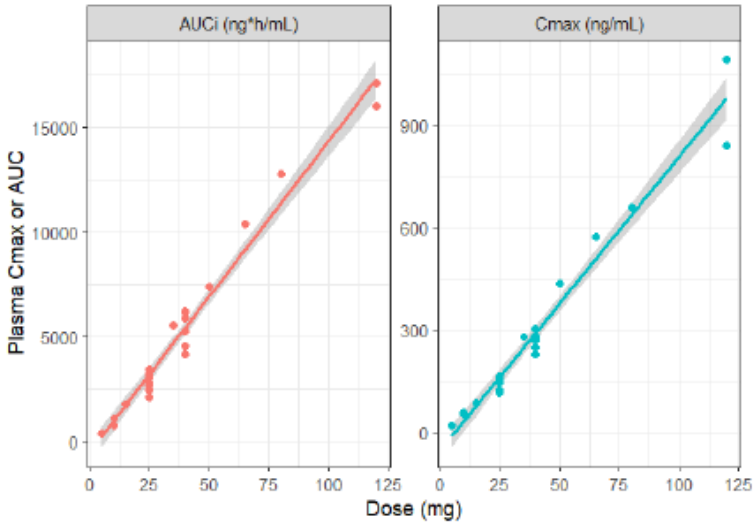
6.2.2.8. Dose proportionality and time dependency

Dose Linearity in Cmax and AUC∞

The Cmax and AUC∞ values over the dose range of 5 to 120 mg after a single oral administration were dose-dependent and linear. The Cmax/D and AUC∞/D were 4.26 to 9.08 ng/mL/mg and 74.8 to 160 ng*h/mL/mg, respectively. At doses of 25 mg or lower, Cmax/D and AUC∞/D were 5.66 ng/mL/mg and 108 ng*h/mL/mg, slightly lower than those at 40 mg of higher doses (7.45 ng/mL/mg and 141 ng*h/mL, respectively). The lower Cmax/D and AUC∞/D at the lower dose region may be explained by

interparticipant variability, the involvement of Pgp in brensocatib absorption, and the transporter saturation status.

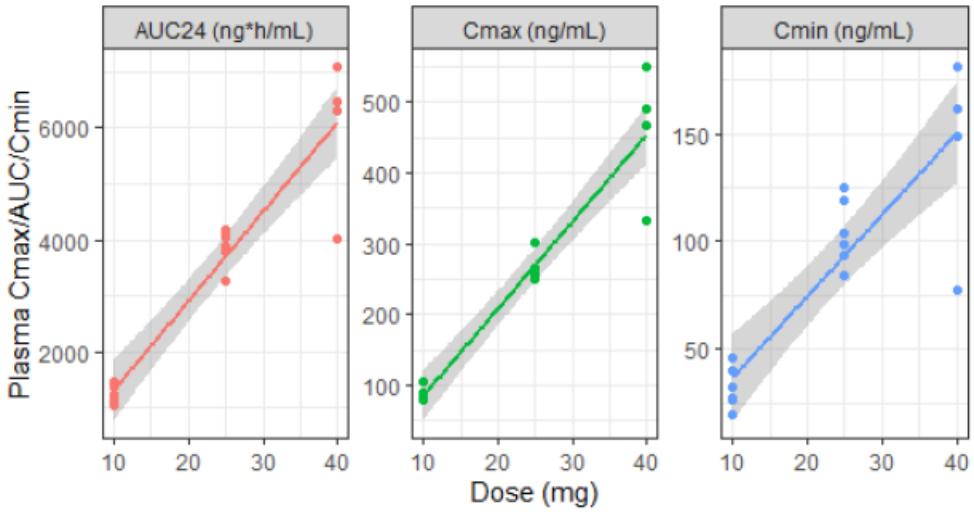
Figure 14: Dose Linearity of Brensocatib Cmax and AUC∞ in Healthy Participants Following a Single Oral Administration at 5 to 120 mg



Source: Module 2.7.1 [Table 16]
Note: Symbols denote group means from individual studies (dots) linear regression model (lines) and its 90% confidence intervals (shaded areas); correlation coefficient (R^2) = 0.965 for C_{max} and 0.973 for AUC_{∞}.

Multiple dose PK with QD dosing was evaluated in 5 studies, including Studies D6190C00001, INS1007-101, INS1007-201, INS1007-211, and INS1007-301. The median Tmax values across these studies (0.75 to 2.9 hours) were similar to that after single dose (0.5 to 2.0 hours).

Figure 15: Steady-state Cmax, AUCtau, and Cmin across studies were generally comparable at the respective dose levels. At 10 to 40 mg, the Cmax and AUCtau showed dose linearity while Cmin data were highly variable.



Source: Section 3 Table 31
Note: Symbols denote group means from individual studies (dots) linear regression model (lines) and its 90% confidence intervals (shaded areas); correlation coefficient (R^2) = 0.889 for AUC_{24} (AUC_{τ}), 0.917 for C_{max}, and 0.779 for C_{min}.

In summary, steady-state brensocatib exposure was dose-dependent and comparable in healthy, NCFBE, and CF participants.

6.2.2.9. Pharmacokinetics in the target population

PK data were obtained in the NCFBE target population from phase 2 study INS1007-201 and phase 3 study INS1007-301. The PK profile of brensocatib in the target population was comparable to that obtained in healthy volunteers. Brensocatib was rapidly absorbed with Tmax around 1 hour. Across the 10 and 25 mg dose, the rate and extent of brensocatib exposure increased with dose. In study INS1007-201, plasma levels (Cmax and AUC) were dose proportional under steady state conditions.

Table 39: Summary of PK Parameters of Brensocatib (PK Substudy, Study INS1007-201)

PK Parameter	Summary Statistics	Brensocatib 10 mg		Brensocatib 25 mg	
		Day 1 (n = 8)	Day 29 (n = 8)	Day 1 (n = 12)	Day 29 (n = 12)
T_{max} (h)	Median (min, max)	0.55 (0.0, 2.0)	2.9 (1.0, 8.0)	1.0 (0.0, 6.0)	1.1 (0.0, 4.0)
C_{max} (ng/mL)	Mean (CV%)	21.2 (110)	80.1 (32.8)	96.6 (79.9)	219 (62.2)
AUC_{0-8} (h*ng/mL)	Mean (CV%)	114 (114)	530 (32.4)	429 (87.1)	1298 (62.5)
AUC_{last} (h*ng/mL)	Mean (CV%)	227 (30.3)	569 (34.5)	687 (27.4)	1513 (52.3)

Source: Study INS1007-201 [Table 14.2.2.2]
Note: Brensocatib was known as INS1007 for Study INS1007-201.

In study INS1007-301, an approximately 2-fold accumulation ratio was observed after multiple dosing for both dose levels (10 mg, 25 mg), while in study INS1007-201 accumulation showed greater variability, however, was in the same order of magnitude (drug accumulation in Cmax and AUCs was 2.5- to 4.6-fold at 10 mg and 2.2- to 3-fold at 25 mg).

In study INS1007-301, PK data were also generated in small subset of adolescent NCFBE participants. Plasma levels in adolescents were generally comparable to those in adult patients.

6.2.2.10. Special populations

Renal Impairment Study INS1007-102

A Phase 1, Open-Label, Single-Dose Parallel-Group Study of Brensocatib Following a Single Oral Administration in Subjects With or Without Renal Impairment

Study INS1007-102 was a Phase 1, open-label, parallel-group study evaluating brensocatib PK, safety, and tolerability in participants with mild, moderate, and severe RI following a single oral administration of 25 mg of brensocatib. Healthy participants matched in sex, age, and BMI served as a control.

Compared to healthy controls, the systemic exposure was similar in the mild, slightly higher in the moderate, but slightly lower in the severe RI participants, without apparent trend regarding the renal functions. Elimination t½, CL/F, and Vd/F were similar across all groups. Statistical evaluation using GMRs showed that brensocatib Cmax was comparable across all groups. The AUC∞ was highly comparable between healthy and the mild groups while the AUCs were 27% higher for the moderate group and 28% lower for the severe group.

Table 40: Comparison of Brensocatib Exposure Between Renal Impairment and Healthy Participants (Study INS1007-102)

Parameter	Cohort	GLSM		GMR (90% CI)
		Participants With Renal Impairment (n)	Healthy Participants (n)	
AUC_{∞} (ng*h/mL)	Mild	3434 (6)	3438 (10)	0.999 (0.776, 1.285)
	Moderate	4297 (6)	3376 (6)	1.273 (0.935, 1.731)
	Severe	2637 (6)	3641 (6)	0.724 (0.543, 0.966)
C_{max} (ng/mL)	Mild	169 (6)	163 (10)	1.033 (0.755, 1.415)
	Moderate	183 (6)	167 (6)	1.096 (0.806, 1.490)
	Severe	139 (6)	167 (6)	0.831 (0.685, 1.008)

Source: Study INS1007-102 [Table 14.2.3]

Hepatic Impairment Study INS1007-105

An Open-label, Phase 1 Study to Evaluate the Pharmacokinetics and Safety of a Single Dose of Brensocatib in Subjects with Normal Hepatic Function and Subjects with Hepatic Impairment

In Study INS1007-105, brensocatib PK and safety were evaluated in participants with mild (Child-Pugh score 5 to 6, n = 6), moderate (Child-Pugh score 7 to 9, n = 6), and severe (Child-Pugh score 10 to 12, n = 6) HI following a single oral administration of 25 mg of brensocatib. Healthy participants (n = 9) served as a control with 6 participants matched to sex, age, and BMI for each of the HI cohorts.

Brensocatib was rapidly absorbed in participants with HI, with a Tmax similar to that in healthy control participants.

The unbound PK parameters were derived based on the percent unbound estimates determined by the plasma protein binding experiments conducted in Study 8496474. The mean fraction of unbound (funb; average of estimates based on 4- and 24-hour samples) was 16.9%, 19.7%, and 26.9% across the HI groups and 17.8% in the normal hepatic function group. The funb was found highly correlated to the baseline serum albumin concentration, suggesting that brensocatib is mainly bound to albumin in the circulation, and the percent unbound appeared to increase with the severity of HI.

Table 41: Statistical Analysis of the Total and Unbound Pharmacokinetic Parameters (Hepatic Impairment Assessment), Study INS1007-105

Parameter	Hepatic Impairment/ Child-Pugh score	n	GLSM	Test Versus Reference Ratio of GLSMs (90% CI)
AUC_{tlast} (ng*h/mL)	Healthy Controls (Reference)	6	2730	
	Child-Pugh 5-6 (Test)	6	3190	1.17 (0.715, 1.90)
	Healthy Controls (Reference)	6	2430	
	Child-Pugh 7-9 (Test)	6	2500	1.03 (0.690, 1.54)
	Healthy Controls (Reference)	6	2410	
	Child-Pugh 10-15 (Test)	6	1930	0.802 (0.495, 1.30)
AUC_{∞} (ng*h/mL)	Healthy Controls (Reference)	6	2780	
	Child-Pugh 5-6 (Test)	6	3330	1.20 (0.732, 1.96)
	Healthy Controls (Reference)	6	2480	
	Child-Pugh 7-9 (Test)	6	2560	1.03 (0.692, 1.54)
	Healthy Controls (Reference)	6	2460	
	Child-Pugh 10-15 (Test)	6	1990	0.810 (0.500, 1.31)
C_{max} (ng/mL)	Healthy Controls (Reference)	6	117	
	Child-Pugh 5-6 (Test)	6	131	1.12 (0.771, 1.63)
	Healthy Controls (Reference)	6	111	
	Child-Pugh 7-9 (Test)	6	106	0.960 (0.670, 1.38)
	Healthy Controls (Reference)	6	105	
	Child-Pugh 10-15 (Test)	6	77.8	0.741 (0.437, 1.26)
$AUC_{tlast,unb}$ (ng*h/mL)	Healthy Controls (Reference)	6	479	
	Child-Pugh 5-6 (Test)	6	539	1.13 (0.774, 1.64)
	Healthy Controls (Reference)	6	428	
	Child-Pugh 7-9 (Test)	6	489	1.14 (0.845, 1.55)
	Healthy Controls (Reference)	6	425	
	Child-Pugh 10-15 (Test)	6	496	1.17 (0.717, 1.90)
$AUC_{\infty,unb}$ (ng*h/mL)	Healthy Controls (Reference)	6	487	
	Child-Pugh 5-6 (Test)	6	564	1.16 (0.793, 1.69)
	Healthy Controls (Reference)	6	437	
	Child-Pugh 7-9 (Test)	6	500	1.14 (0.847, 1.55)
	Healthy Controls (Reference)	6	435	
	Child-Pugh 10-15 (Test)	6	511	1.18 (0.723, 1.91)
$C_{max,unb}$ (ng/mL)	Healthy Controls (Reference)	6	20.4	
	Child-Pugh 5-6 (Test)	6	22.1	1.08 (0.803, 1.46)
	Healthy Controls (Reference)	6	19.5	
	Child-Pugh 7-9 (Test)	6	20.8	1.07 (0.767, 1.48)
	Healthy Controls (Reference)	6	18.6	
	Child-Pugh 10-15 (Test)	6	20.0	1.08 (0.710, 1.63)

Data were analyzed using a separate analysis of variance model for each hepatic impairment group vs normal controls (group main effect; 1 df)
The comparison reference group (ie, normal hepatic function group) only included those subjects who were matched to a subject within each specific test group
The GLSMs, ratio of GLSMs, and corresponding CIs were obtained by taking the exponential of the LSMs, differences in LSMs, and corresponding CIs on the ln scale.
Abbreviations: AUC_{tlast} = area under the concentration-time curve from time 0 to the time of the last quantifiable concentration (t_{last}); AUC_{∞} = area under the concentration-time curve from time 0 extrapolated to infinity; CI = confidence interval; C_{max} = maximum observed concentration; $AUC_{tlast,unb}$ = unbound AUC_{tlast}, calculated as $AUC_{tlast}*f_{unb}$; $AUC_{\infty,unb}$ = unbound AUC_{∞}, calculated as $AUC_{\infty}*f_{unb}$; $C_{max,unb}$ = unbound C_{max}, calculated as $C_{max}*f_{unb}$; f_{unb} = fraction unbound; GLSM = geometric least squares mean; ln = natural log; LSM = least square mean; n = number of subjects with valid observations; 'unb' stands for unbound parameters.
Reference: Table 14.2.1.6

All unbound PK parameters were comparable across all groups, ranging from 490 to 554 h*ng/mL for unbound AUCtlast (AUCtlast,unb); 500 to 572 h*ng/mL for unbound AUC∞ (AUC∞,unb); and 21.0 to 22.5 ng/mL for unbound Cmax (Cmax,unb). The data indicated that although total drug exposure varied among groups, the unbound drug exposure remained unaffected.

6.2.2.11. Pharmacokinetic interaction studies

In vitro data indicated that brensocatib is a substrate of CYP3A, Pgp, and BCRP.

DDI Study D6190C00003 of brensocatib with verapamil, itraconazole, diltiazem

An open-label, non-randomised, fixed sequence study assessing the pharmacokinetics of INS1007 when administered alone and with multiple doses of verapamil and itraconazole or diltiazem in healthy subjects

This was a Phase I, open-label, non-randomised, fixed sequence, 3-period study in 15 healthy male subjects at a single study centre.

Cytochrome P450 (CYP) enzymes CYP3A4 and CYP3A5 are the predominant CYP isoforms involved in the metabolism of INS1007 in vitro. The study was designed to assess the PK of INS1007 in healthy subjects when administered alone and in combination with multiple doses of verapamil and itraconazole or diltiazem and sized to test for bioequivalence with these drugs. Verapamil and diltiazem are moderate CYP3A4 inhibitors, whereas itraconazole is a potent CYP3A4 inhibitor.

After co-administration of brensocatic with CYP3A4 inhibitor verapamil, the exposure of brensocatic was moderately increased (AUC by about 33% and Cmax by 53%). It was surprising that itraconazole had a smaller effect than verapamil on brensocatib's AUC (increased by only 12-13%), – since itraconazole is a stronger CYP3A4 inhibitor than verapamil. This indicates that clearance routes other than metabolism by CYP3A4 are also important in the clearance of INS1007. The reason that verapamil had a bigger effect than itraconazole may be related to factors other than CYP3A4 inhibition which was not fully understood at the same when Study D6190C100003's CSR was edited. However, based on the changes in PK parameters of INS1007 observed, the phenomenon could be related to changes in the fraction of INS1007 absorbed.

Table 42: Geometric Least Square Mean, Geometric Least Square Mean 90% CIs, Pairwise Comparison, Ratio of INS1007 Following Single Administration of INS1007, and in Combination with Verapamil or Itraconazole to Healthy Subjects under Fasted Conditions, Study D6190C00003

Pharmacokinetic parameter (Unit)	Treatment	n	Geometric LS Mean	95% CI	Pairwise Comparison		
					Pair	Ratio	90% CI
C_{max} (nmol/L)	AZD7986	15	385.8	[327.8, 454.1]			
	AZD7986 + verapamil	15	591.9	[502.9, 696.6]	AZD7986 + verapamil / AZD7986	153.40	[136.16, 172.83]
	AZD7986 + itraconazole	15	234.1	[198.9, 275.5]	AZD7986 + itraconazole / AZD7986	60.66	[53.84, 68.34]
AUC_{last} (h*nmol/L)	AZD7986	15	6545	[5311, 8066]			
	AZD7986 + verapamil	15	8739	[7091, 10770]	AZD7986 + verapamil / AZD7986	133.51	[122.70, 145.29]
	AZD7986 + itraconazole	15	7361	[5973, 9071]	AZD7986 + itraconazole / AZD7986	112.45	[103.34, 122.37]
AUC (h*nmol/L)	AZD7986	15	6697	[5431, 8258]			
	AZD7986 + verapamil	15	8857	[7183, 10920]	AZD7986 + verapamil / AZD7986	132.25	[121.78, 143.64]
	AZD7986 + itraconazole	15	7615	[6175, 9390]	AZD7986 + itraconazole / AZD7986	113.70	[104.69, 123.49]

CI: confidence interval; LS: least-squares; N: all subjects in the Pharmacokinetic analysis set who received treatment; n: all subjects included in the statistical comparison analysis. Result based on analysis of variance (ANOVA) of log-transformed PK parameter with treatment as fixed effect and subject as random effect. Geometric mean ratio and CI were back-transformed and presented as percentages. Geometric LS mean and 95% CI were also back-transformed.

Data Source: End-of-Text Table 14.2.3.

DDI Study INS1007-109 of Brensocatib with Clarithromycin

A Phase 1, Open-label, Fixed-sequence Study to Investigate the Effect of Clarithromycin, a Strong CYP3A4 Inhibitor, on Brensocatib Pharmacokinetics in Healthy Subjects

This was an open-label, nonrandomized, fixed-sequence study in healthy females of nonchildbearing potential and male subjects to evaluate the impact of the strong CYP3A and P-gp inhibitor, clarithromycin, on brensocatib PK, safety, and tolerability.

Brensocatib metabolism is mainly through CYP3A4/5. It is also a substrate of P-gp. Clarithromycin is an FDA-recommended strong CYP3A4 and P-gp inhibitor that has been previously used in several clinical

studies. Based on a review of clinical, in vitro, and modelling data, 5 days of continuous dosing appears to reach the highest level of CYP3A4 inhibition.

The PK parameters of brensocatib administered alone were consistent with those observed in previous studies. When brensocatib was co-administered with clarithromycin, a CYP3A4 and P-gp inhibitor, the ratios of GLSMs (90% CI) for AUCtlast, AUCinf, and Cmax of brensocatib were 1.558 (1.456, 1.668), 1.555 (1.455, 1.662) and 1.675 (1.501, 1.870), respectively, compared to brensocatib administered alone, and t1/2 was delayed by approximately 4 hours (arithmetic mean t1/2 values of 29.0 hours compared to 25.0 hours). The statistical significance of these results was indicated through the 90% CIs of the ratios of GLSMs falling outside of the prespecified boundaries of 0.80 to 1.25, suggesting that clarithromycin significantly increased the extent and rate of brensocatib exposure and reduced the rate of elimination (t1/2).

Table 43: Statistical Analysis of Pharmacokinetic Parameters (Drug-drug Interaction Assessment) Following a Single Dose of Brensocatib Alone or a Single Dose of Brensocatib in the Presence of Steady-state Exposure of Clarithromycin, Study INS1007-109

				Test versus Reference	
Parameter	Treatment	n	GLSM	Ratio of GLSMs (90% CI)	Within-subject CV
AUC_{tlast} (h*ng/mL)	25 mg Brensocatib (Reference)	22	2390	---	---
	25 mg Brensocatib + 500 mg Clarithromycin BID (Test)	22	3720	1.558 (1.456, 1.668)	13.1
AUC_{inf} (h*ng/mL)	25 mg Brensocatib (Reference)	22	2440	---	---
	25 mg Brensocatib + 500 mg Clarithromycin BID (Test)	22	3800	1.555 (1.455, 1.662)	12.9
C_{max} (ng/mL)	25 mg Brensocatib (Reference)	22	124	---	---
	25 mg Brensocatib + 500 mg Clarithromycin BID (Test)	22	207	1.675 (1.501, 1.870)	21.4
$t_{1/2}$ (h)#	25 mg Brensocatib (Reference)	22	24.8	---	---
	25 mg Brensocatib + 500 mg Clarithromycin BID (Test)	22	28.2	4.118 (2.486, 5.402)	---

The n, median, and Hodges-Lehmann estimate of median difference (90% CI) from the Wilcoxon signed-rank test presented.
Model: ln(parameter) = treatment + subject + random error, with subject fitted as a random effect
The GLSMs, ratios of GLSMs, and corresponding CIs were obtained by taking the exponential of the LSMs, differences in LSMs, and corresponding CIs on the ln scale.
Abbreviations: AUC_{inf} = area under the concentration-time curve from time 0 extrapolated to infinity; AUC_{tlast} = area under the concentration-time curve from time 0 to the time of the last quantifiable concentration; BID = twice daily; CI = confidence interval; C_{max} = maximum observed concentration; CV = coefficient of variation; GLSM = geometric least squares mean; ln = natural log; LSM = least squares mean; n = number of subjects; $t_{1/2}$ = apparent terminal elimination half-life
Reference: Table 14.2.1.3

DDI Study INS1007-106 of Brensocatib with rifampin and esomepromazole

A Phase 1, Open label, Fixed Sequence Study to Assess the Pharmacokinetics of Brensocatib when Administered Alone and With Multiple Doses of Rifampin (CYP3A Inducer) or Esomeprazole (Proton Pump Inhibitor) in Healthy Subjects

This study was an open label, fixed sequence, 2-part, 2-period study to determine the PK, safety, and tolerability of brensocatib when administered alone and in combination with multiple doses of rifampin (Part 1) or esomeprazole (Part 2) in healthy male and female subjects to evaluate the impact of CYP3A induction on brensocatib PK and gastric pH elevation on brensocatib oral absorption.

Brensocatib is a substrate of CYP3A, P-gp and BCRP. Rifampin is a potent inducer of CYP3A, and other CYP isozymes. Rifampin is also a known P-gp inducer and activates P-gp in the gastrointestinal wall which increases the intestinal secretion of victim drugs. Once daily administration of rifampin at 600 mg for 7 to 10 days was intended to achieve maximum induction effect on the activity of CYP3A and P-gp.

Brensocatib shows pH dependent solubility. Elevation of gastric pH by acid-reducing agents (ARAs) can affect the solubility and dissolution characteristics of orally administered brensocatib. As a result, concomitant administration of an ARA drug may potentially decrease the oral absorption of brensocatib. In this study, esomeprazole, a known PPI and recommended by FDA Guidance, was used to characterize the potential for a pH dependent DDI.

Based on the statistical analysis and the ratios of GLSMs, the Cmax, AUCtlast, AUCinf, and elimination t1/2 of brensocatib (co-administered with rifampin) reduced by 15%, 33%, 33%, and 26%, respectively, compared to when brensocatib was dosed alone. The statistical significance of these results was indicated through the 90% CIs for the ratios of GLSMs for Cmax, AUCtlast, AUCinf, and elimination t1/2 falling outside the bioequivalence boundaries (0.80, 1.25).

Table 44: Statistical Analysis of Pharmacokinetic Parameters (Drug-drug Interaction Assessment) Following a Single Dose of Brensocatib Alone or a Single Dose of Brensocatib in the Presence of Steady-state Exposure of Rifampin, Study INS1007-106

				Test versus Reference	
Parameter	Treatment	n	GLSM	Ratio of GLSMs (90% CI)	Within-subject CV%
AUC_{tlast} (h*ng/mL)	25 mg brensocatib (Reference)	16	3150	---	---
	25 mg brensocatib + 600 mg rifampin (Test)	14	2110	0.671 (0.606, 0.743)	15.4
AUC_{inf} (h*ng/mL)	25 mg brensocatib (Reference)	16	3240	---	---
	25 mg brensocatib + 600 mg rifampin (Test)	14	2170	0.670 (0.606, 0.741)	15.1
C_{max} (ng/mL)	25 mg brensocatib (Reference)	16	154	---	---
	25 mg brensocatib + 600 mg rifampin (Test)	14	130	0.846 (0.763, 0.938)	15.5
$t_{1/2}$ (h)	25 mg brensocatib (Reference)	16	27.0	---	---
	25 mg brensocatib + 600 mg rifampin (Test)	14	19.9	0.738 (0.675, 0.806)	13.4
$t_{1/2}$ (h)#	25 mg brensocatib (Reference)	14	27.2	---	---
	25 mg brensocatib + 600 mg rifampin (Test)	14	22.1	-6.38 (-7.85, -5.13)	---

The n, median, and Hodges-Lehmann estimate of median difference (90% CI).
Model: ln(parameter) = treatment + random error, with subject fitted as a random effect
The GLSMs, ratios of GLSMs, and corresponding CIs were obtained by taking the exponential of the LSMs, differences in LSMs, and corresponding CIs on the ln scale.
Abbreviations: AUC_{inf} = area under the concentration-time curve from time 0 extrapolated to infinity; AUC_{tlast} = area under the concentration-time curve from time 0 to the time of the last quantifiable concentration; CI = confidence interval; C_{max} = maximum observed concentration; CV = coefficient of variation (%); GLSM = geometric least squares mean; ln = natural log; LSM = least square mean; n = number of subjects with valid observations; $t_{1/2}$ = apparent terminal elimination half-life
Reference: Table 14.2.1.3a

Co-administration of brensocatib with PPI esomepromazole did not affect brensocatib exposure. Based on the statistical analysis, the 90% CIs for the ratios of GLSMs for Cmax, AUCtlast, and AUCinf were completely within the bioequivalence boundaries (0.959 [0.868, 1.06], 1.05 [1.01, 1.08], and 1.05 [1.01, 1.09], respectively) when brensocatib was administered with esomeprazole compared to when administered alone.

Table 45: Statistical Analysis of Pharmacokinetic Parameters (Drug-drug Interaction Assessment) Following a Single Dose of Brensocatib Alone and in the Presence of Steady-state Exposure of Esomeprazole, Study INS1007-106

				Test versus Reference	
Parameter	Treatment	n	GLSM	Ratio of GLSMs (90% CI)	Within-subject CV%
AUC_{tlast} (h*ng/mL)	25 mg brensocatib (Reference)	16	2990	---	---
	25 mg brensocatib + 40 mg esomeprazole (Test)	16	3140	1.05 (1.01, 1.08)	5.51
AUC_{inf} (h*ng/mL)	25 mg brensocatib (Reference)	16	3050	---	---
	25 mg brensocatib + 40 mg esomeprazole (Test)	16	3210	1.05 (1.01, 1.09)	5.64
C_{max} (ng/mL)	25 mg brensocatib (Reference)	16	154	---	---
	25 mg brensocatib + 40 mg esomeprazole (Test)	16	148	0.959 (0.868, 1.06)	16.2

Model: ln(parameter) = treatment + random error, with subject fitted as a random effect
The GLSMs, ratios of GLSMs, and corresponding CIs were obtained by taking the exponential of the LSMs, differences in LSMs, and corresponding CIs on the ln scale.
Abbreviations: AUC_{inf} = area under the concentration-time curve from time 0 extrapolated to infinity; AUC_{tlast} = area under the concentration-time curve from time 0 to the time of the last quantifiable concentration; CI = confidence interval; C_{max} = maximum observed concentration; CV = coefficient of variation (%); GLSM = geometric least squares mean; ln = natural log; LSM = least square mean; n = number of subjects with valid observations
Reference: Table 14.2.1.3b

6.2.3. Pharmacodynamics

6.2.3.1. Mechanism of action

Brensocatib is an oral, selective, competitive, and reversible inhibitor of dipeptidyl peptidase 1 (DPP1). DPP1 activates pro inflammatory neutrophil serine proteases (NSPs) during neutrophil maturation in the bone marrow. Activated NSPs are implicated in the pathogenesis of many neutrophil-mediated inflammatory diseases, including bronchiectasis. Brensocatib reduces the activity of NSPs including neutrophil elastase, cathepsin G and proteinase 3.

6.2.3.2. Primary and secondary pharmacology

6.2.3.2.1. Primary pharmacology

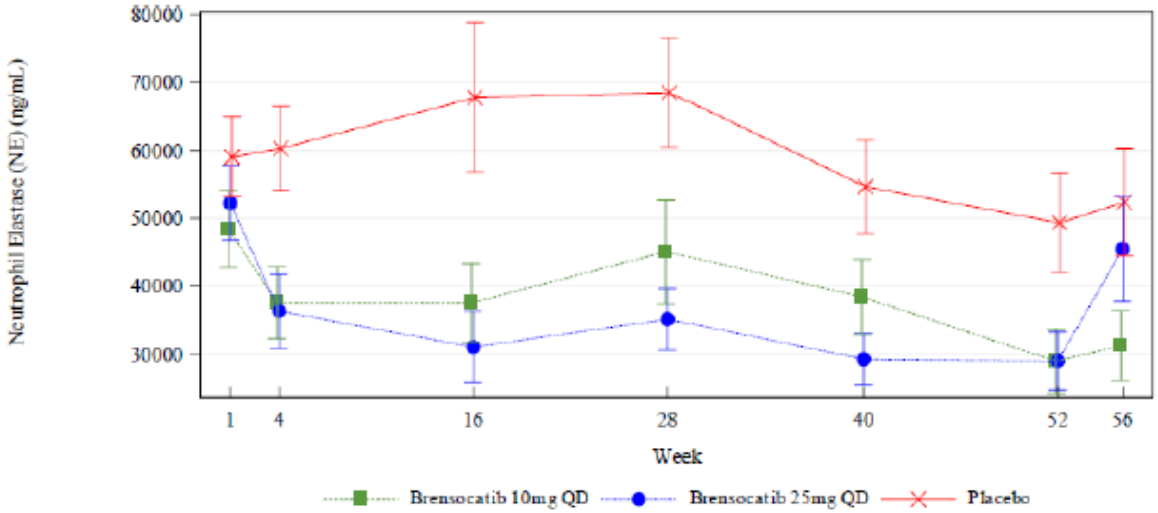
Pharmacodynamic Biomarkers

The activity of NSP enzymes (NE, CatG, and PR3) was evaluated following QD dosing in Studies D6190C00001 (NE in plasma only), INS1007-201 (blood and sputum), INS1007-211 (blood and sputum), and INS1007-301 (blood and sputum). Regardless of the variability of the NSP data, a dose- and exposure-dependent reduction in the concentrations of active NSP in blood and sputum was observed.

Subjects with Cystic Fibrosis (Study 211) and NCFBE (Studies 201 and 301)

Following QD administration of brensocatib in participants with NCFBE (24 or 52 weeks) and CF (4 weeks), the maximum reduction in NSP activity was attained generally around 4 weeks, maintained relatively constant until end of treatment and then recovered at the follow-up visit (4 weeks after the last dose).

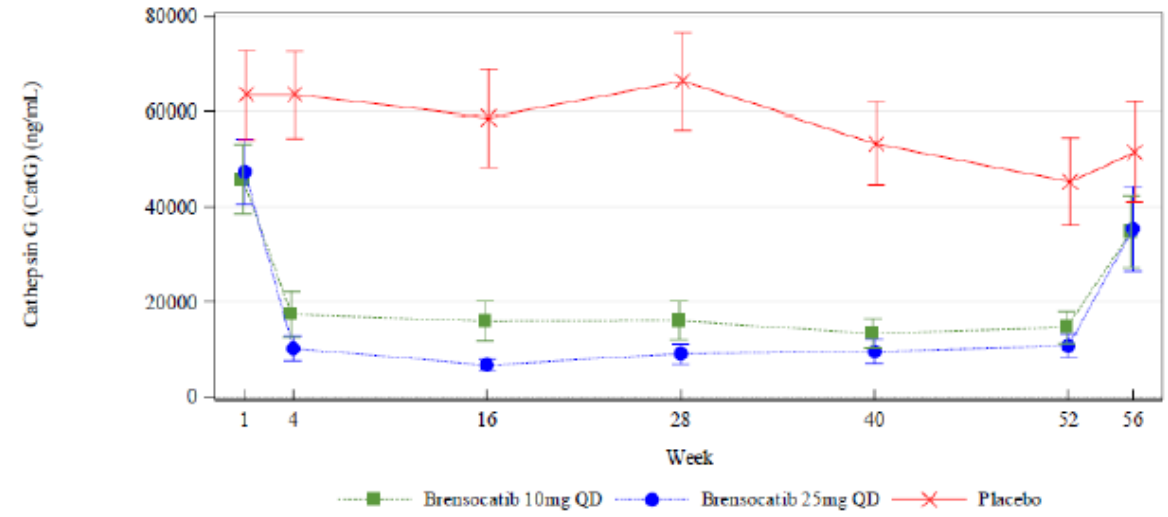
Figure 16: Mean (± SE) Concentrations of Active Neutrophil Elastase in Sputum –Adult Participants (PD Analysis Set), Phase 3 Study INS1007-301



Number of Participants with Observation

Brensocatib 10mg QD	105	96	97	92	88	81	69
Brensocatib 25mg QD	108	96	98	95	87	85	67
Placebo	115	102	102	104	100	83	77

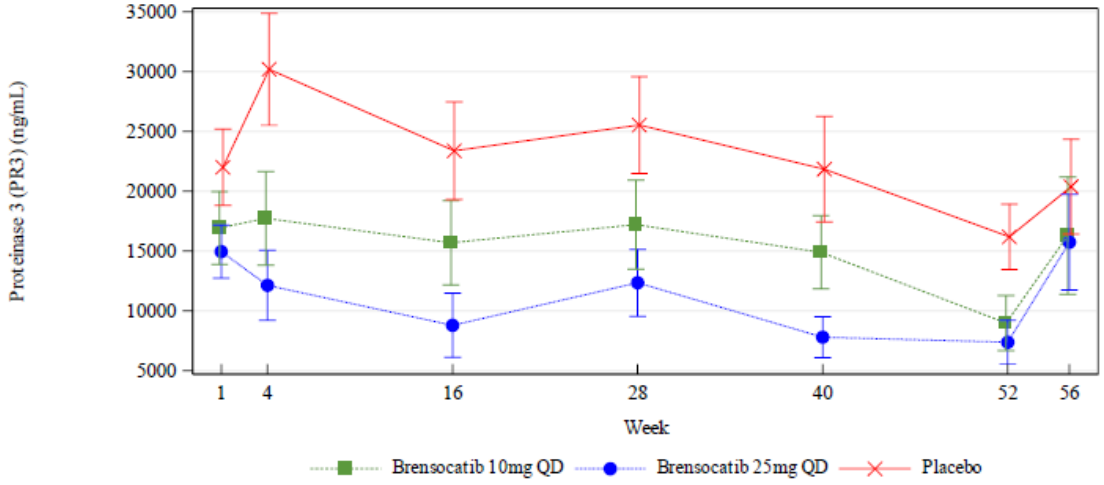
Figure 17: Mean (± SE) Concentrations of Active Cathepsin G in Sputum –Adult Participants (PD Analysis Set), Phase 3 Study INS1007-301



Number of Participants with Observation

Brensocatib 10mg QD	105	95	97	92	88	81	69
Brensocatib 25mg QD	108	96	98	95	87	84	67
Placebo	115	102	102	104	100	83	77

Figure 18: Mean (± SE) Concentrations of Active Proteinase 3 in Sputum –Adult Participants (PD Analysis Set), Phase 3 Study INS1007-301



Number of Participants with Observation

Brensocatib 10mg QD	105	96	97	91	88	81	69
Brensocatib 25mg QD	108	96	97	95	87	85	67
Placebo	115	102	102	104	100	83	77

Source: Figure 16.2.10.1-2i
An On-Investigation Product approach was used for handling of intercurrent events.
Pretreatment (baseline) at Week 1 was the arithmetic mean value of pharmacodynamic parameter results at Screening and Day 1.
QD = once daily, SE = standard error

6.2.3.2.2. Secondary pharmacology

QT Study INS1007-104

A Two-part, Phase I, Double-blind, Placebo- and Positive-controlled Crossover Study to Investigate the Effects of Brensocatib on QT Interval in Healthy Subjects

This study evaluated the potential effects of Brensocatib on cardiac repolarization, specifically focusing on the QT interval corrected using Fridericia's formula (QTcF). The study was conducted in two parts: the first aimed to identify a safe supratherapeutic dose, and the second assessed the QTc impact of therapeutic and supratherapeutic doses of Brensocatib in a randomized, double-blind, placebo- and positive-controlled crossover design.

The data presented demonstrate that Brensocatib, when administered as single oral doses of 40 mg (therapeutic) and 120 mg (supratherapeutic), does not have a clinically meaningful effect on QTc interval prolongation. The observed QTc changes remained well within acceptable regulatory limits, with a maximal placebo-corrected change (ΔΔQTcF) below the 10 ms threshold of concern, even at peak plasma concentrations.

The use of moxifloxacin as a positive control confirmed the assay sensitivity, supporting the robustness and validity of the study's design and analytical methods. Brensocatib exhibited predictable, dose-proportional pharmacokinetics and was well tolerated, with only mild, transient adverse events reported and no clinically significant findings in ECGs, laboratory parameters, or vital signs.

Overall, the thorough QT assessment supports the cardiac safety profile of Brensocatib at both therapeutic and supratherapeutic doses. These results align with ICH E14 guidance for negative TQT studies.

6.2.3.3. Pharmacodynamic interactions with other medicinal products or substances

The issue of potential pharmacodynamic DDI was not explored.

6.2.3.4. Genetic differences in PD response

The issue of genetic differences in PD response was not addressed.

6.2.3.5. Immunological events

N/A

6.2.4. Pharmacokinetics/pharmacodynamics (PK/PD)

Report 00566-1

Data for PK-PD analyses came from Study INS1007-201 only. A total of 241 subjects from the PD population were available for the PK-PD analyses; 157 subjects who received INS1007 and had estimates of INS1007 exposure from the population PK analysis (75 received 10 mg QD, 82 received 25 mg QD) and 84 subjects who received placebo.

Table 46: Description of PK-PD analysis populations

Outcome evaluated	N	Comment
Pulmonary Exacerbation (dichotomous)	241	All subjects in PD population per Insmed; Subjects randomized to INS1007 were required to have estimates of INS1007 exposure from the population PK analysis
Pulmonary Exacerbation (time-to-event)	241	
Sputum NE	237	Excluding subjects without sputum NE observations (n = 4; 2 from INS1007 25 mg QD; 2 from placebo)
Sputum NE BLQ post-baseline	237	
Correlation of pulmonary exacerbation wit sputum NE	237	
AESI	242	All subjects in safety population per Insmed; Subjects randomized to INS1007 were required to have estimates of INS1007 exposure from the population PK analysis

Note: Abbreviations are provided in the Abbreviation Listing.

Two efficacy outcome variables were used as the basis for the PK-PD analyses for efficacy:
1) pulmonary exacerbations and
2) concentrations of active neutrophil elastase (NE) in sputum

Pulmonary exacerbation was evaluated in two ways:
1) dichotomous (yes or no for occurrence of a pulmonary exacerbation prior to Week 24) and
2) using time-to-event models (time to occurrence of a pulmonary exacerbation).

Sputum samples for the determination of NE activity were to be obtained at screening (Day -42 to -1), baseline (Day 1), and Weeks 2, 4, 12, 24, and 28 (follow-up). Sputum NE was evaluated as a continuous variable (e.g., absolute values or change over time and maximum change from baseline) and as a dichotomous variable [achievement of a value below the limit of quantitation (BLQ) post-baseline]. Post-baseline BLQ values that were achieved after the occurrence of a pulmonary exacerbation were excluded due to the fact that the administration of antibiotics for an exacerbation have the potentially to suppress sputum NE activity.

Adverse events of special interest (AESI) to be evaluated included the following: periodontal disease, hyperkeratosis, and the occurrence of "other" infections (i.e, not pulmonary exacerbations).

Efficacy analysis

Pulmonary Exacerbations

The incidence of pulmonary exacerbations by steady-state AUC0-24 quartile are provided in table below.

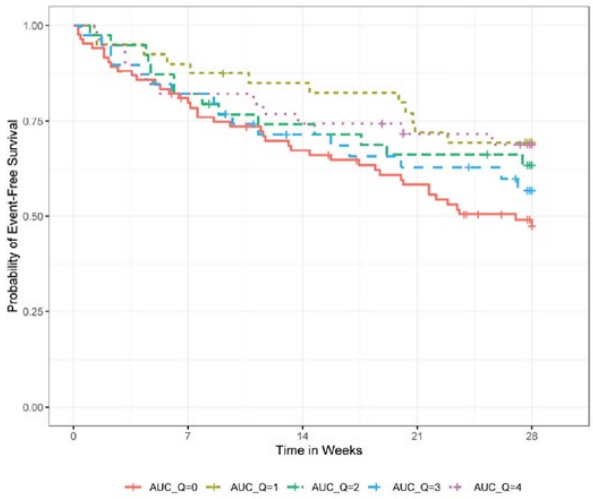
Table 47: Incidence of pulmonary exacerbation by AUC0-24 quartile

Quartile (AUC_{0-24} range in ng•h/mL)	Incidence of pulmonary exacerbation
Placebo	42/84 (50.0%)
Q1 (624 - 1758)	12/40 (30.0%)
Q2 (1813 - 2754)	14/39 (35.9%)
Q3 (2840 - 4824)	16/39 (41.0%)
Q4 (4845 - 10562)	12/39 (30.8%)

Note: Abbreviations are provided in the Abbreviation Listing.

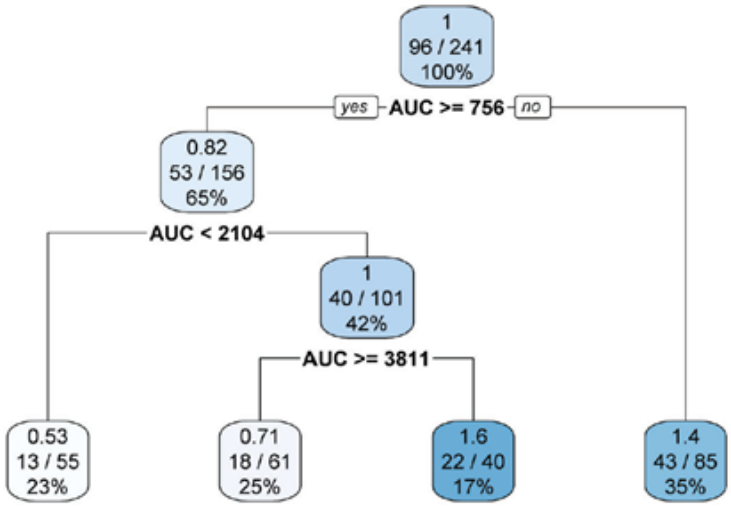
Kaplan-Meier curves showing the time to pulmonary exacerbation stratified by AUC quartile are provided in figure below.

Figure 19: Kaplan-Meier curves showing the time to pulmonary exacerbation, stratified by steady-state INS1007 AUC0-24 quartile



Classification and regression tree plot was used to identify thresholds for AUC0-24 that were predictive of differences in the time to occurrence of pulmonary exacerbations by a Cox Proportional hazards model as shown in figure below.

Figure 20: Classification-and-regression tree plot showing the relationship between pulmonary exacerbations and steady-state INS1007 AUC0-24



Note: Abbreviations are provided in the Abbreviation Listing. Values in the boxes represent the hazard ratio for incidence of pulmonary exacerbation in the corresponding bin (top number), the pulmonary exacerbation rate in the bin (middle numbers), and the percentage of subjects in the bin (lower percentage).

Sputum Neutrophil Elastase

Potential relationships between the occurrence of a sputum NE BLQ observation post-baseline was observed when steady-state INS1007 AUC0-24 was evaluated as an independent predictor. As show in table below subjects treated with INS1007 were more likely to achieve sputum NE BLQ post-baseline than those randomized to placebo and subjects in the highest AUC0-24 quartile had the highest incidence of sputum NE BLQ post-baseline.

Table 48: Incidence of BLQ post-baseline in sputum NE by AUC0-24 quartile

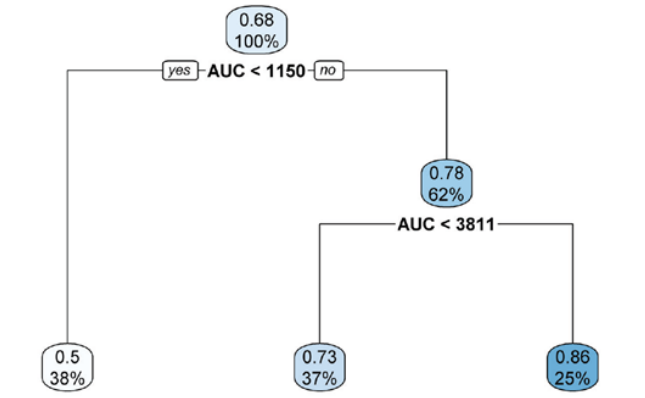
Quartile (AUC_{0-24} range in ng•h/mL)	Incidence of Sputum NE BLQ Post-Baseline
Placebo	41/82 (50.0%)
Q1 (624 - 1758)	31/40 (77.5%)
Q2 (1813 - 2753)	28/39 (71.8%)
Q3 (2840 - 4824)	27/38 (71.1%)
Q4 (4845 - 9466)	33/38 (86.8%)

Note: Abbreviations are provided in the Abbreviation Listing.

Attempts to fit logistic regression models with sputum NE BLQ post-baseline as the dependent variable and steady-state AUC0-24 as the independent predictor were not discriminatory as the model fit to the data from all subjects resulted in significant differences between active and placebo subjects only and the model fit to the data from subjects receiving active INS1007 only showed no significant relationships.

Two thresholds were found in the distribution of AUC0-24 using CART (AUC0-24 of 1150 and 3811) at which the incidence of sputum NE BLQ post-baseline was substantially different above and below the thresholds.

Figure 21: Classification-and-regression tree plot showing the relationship between sputum NE BLQ post-baseline and steady-state INS1007 AUC0-24



Note: Abbreviations are provided in the Abbreviation Listing. Values in the boxes represent the incidence of sputum NE BLQ post-baseline in the corresponding bin (top number) and the percentage of subjects in the bin (lower percentage).

Table 49: Incidence of BLQ post-baseline in sputum NE by CART-Derived AUC0-24 threshold (Subjects Randomized to INS1007 Only)

AUC_{0-24} range in ng•h/mL	Incidence of Sputum NE BLQ Post-Baseline
< 1150	4/8 (50.0%)
1150-3810	64/88 (72.7%)
> 3810	51/59 (86.4%)

Note: Abbreviations are provided in the Abbreviation Listing.

Sputum Neutrophil Elastase as Predictive of Pulmonary Exacerbations

In order to evaluate the predictive value of sputum NE as a marker for pulmonary exacerbations, exploratory analyses were conducted comparing changes in sputum NE to the occurrence of pulmonary exacerbations.

There was a slight trend for larger decreases in sputum NE in subjects who did not experience a pulmonary exacerbation as compared to those who did experience a pulmonary exacerbation during the 24-week study period. However, the difference is small (-98% vs -90%) and the variability in sputum NE change during the study period resulted in a lack of statistical significance.

In contrast, when evaluated as a categorical variable based on achievement of sputum NE BLQ post-baseline, there were clear trends for subjects who achieved sputum NE BLQ post-baseline to have a lower incidence of pulmonary exacerbations.

Table 50: Two-by-two tables showing the incidence of pulmonary exacerbations, stratified by achievement of sputum NE BLQ post-baseline

Study Population	Sputum NE BLQ Post-Baseline?	Pulmonary Exacerbation	
		Yes	No
All subjects[a]	Yes	47	113
	No	48	29
Active INS1007[b]	Yes	32	87
	No	22	14

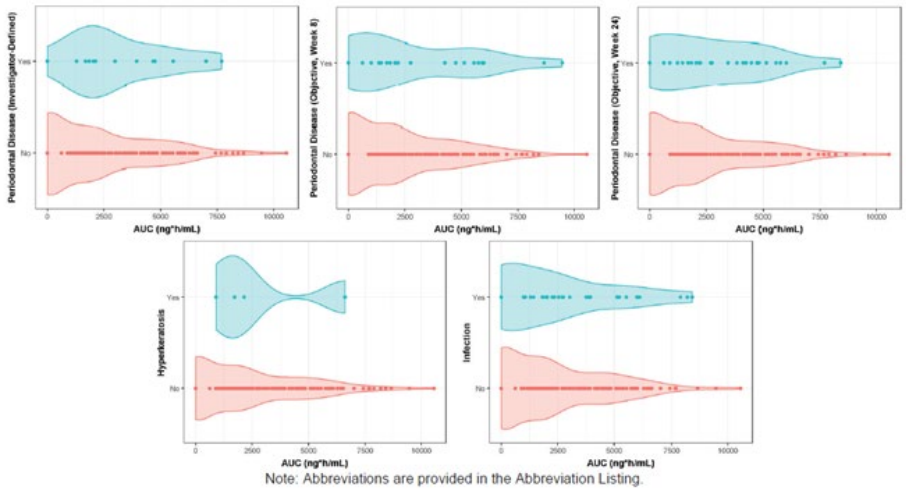
Note: Abbreviations are provided in the Abbreviation Listing.
a. PPV = 0.706; NPV = 0.623
b. PPV = 0.731; NPV = 0.611

The trends are significant based on the log-rank test applied to the time-to-event dana regardless of which population is used. The hazard ratios (with associated 95% confidence intervals) in subjects who achieved sputum NE BLQ post-baseline relative to those that did not were 0.32 (0.21 – 0.48) and 0.29 (0.17 – 0.51) in all subjects and subjects randomized to INS1007 only, respectively.

Safety analysis

The PK-PD analyses for safety did not reveal any clinically-relevant relationships. The exploratory screening plots for the AESI by INS1007 AUC0-24 are provided in figure below.

Figure 22: Horizontal violin plots showing the distributions of steady-state AUC0-24 by occurrence of AESI



Note: Abbreviations are provided in the Abbreviation Listing.

Similarly, evaluation of the incidence AESI by AUC0-24 quartile failed to show substantial differences across the range of INS1007 exposure in subjects randomized to INS1007.

Table 51: Incidence of AESI by steady-state AUC0-24 quartile

AESI	AUC_{0-24} quartile	No	Yes
Periodontal disease (investigator-defined)	Placebo	83 (97.6)	2 (2.35)
	Q1	38 (95.0)	2 (5.00)
	Q2	33 (84.6)	6 (15.4)
	Q3	35 (89.7)	4 (10.3)
	Q4	36 (92.3)	3 (7.69)
Periodontal disease (objective-defined, Week 8)	Placebo	76 (89.4)	9 (10.6)
	Q1	35 (87.5)	5 (12.5)
	Q2	34 (87.2)	5 (12.8)
	Q3	37 (94.9)	2 (5.13)
	Q4	31 (79.5)	8 (20.5)
Periodontal disease (objective-defined, Week 24)	Placebo	76 (89.4)	9 (10.6)
	Q1	35 (87.5)	5 (12.5)
	Q2	32 (82.1)	7 (17.9)
	Q3	32 (82.1)	7 (17.9)
	Q4	33 (84.6)	6 (15.4)
Hyperkeratosis	Placebo	85 (100)	NA
	Q1	38 (95.0)	2 (5.00)
	Q2	38 (97.4)	1 (2.56)
	Q3	39 (100)	NA
	Q4	38 (97.4)	1 (2.56)
Infection	Placebo	69 (81.2)	16 (18.8)
	Q1	36 (90.0)	4 (10.0)
	Q2	29 (74.4)	10 (25.6)
	Q3	35 (89.7)	4 (10.3)
	Q4	30 (76.9)	9 (23.1)

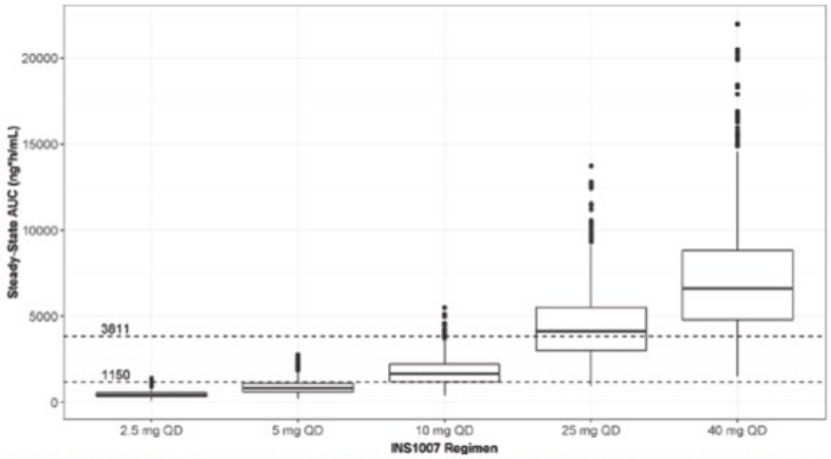
Note: Abbreviations are provided in the Abbreviation Listing.

Model-Based Simulations

INS1007 Pharmacokinetic Exposure Predictions by Dose

A comparison of the predicted steady-state AUC0-24 across the simulated dose regimens are provided in figure below.

Figure 23: Box-and-whisker plots showing the distribution of predicted steady-state AUC0-24 by dose regimen in hypothetical patient population



Note: Abbreviations are provided in the Abbreviation Listing. Horizontal dashed lines show the AUC_{0-24} thresholds for achievement of sputum NE BLQ (**Section 4.2.2.2**)

As expected, the probability of achieving INS1007 exposures that are associated with higher likelihood of achieving sputum NE BLQ post-baseline increases with increasing dose.

Predicted Outcomes by Dose

The predicted incidences of achievement of sputum NE BLQ post-baseline and pulmonary exacerbations based upon the model-based simulations are provided in table below.

Table 52: Median (90% prediction interval) predicted outcomes by simulated dosing regimen based upon the model-based simulations

Outcome	Placebo	INS1007 Dose (mg QD)				
		2.5	5	10	25	40
Incidence of sputum NE BLQ post-baseline	0.496 (0.408 - 0.594)	0.498 (0.409 - 0.595)	0.549 (0.481 - 0.628)	0.682 (0.618 - 0.747)	0.806 (0.750 - 0.865)	0.850 (0.779 - 0.913)
Delta from placebo		0.0020 (-0.001 - 0.006)	0.051 (0.023 - 0.084)	0.178 (0.0909 - 0.279)	0.306 (0.205 - 0.409)	0.348 (0.238 - 0.458)
Incidence of pulmonary event by Week 24	0.440 (0.358 - 0.525)	0.439 (0.359 - 0.523)	0.424 (0.347 - 0.499)	0.380 (0.307 - 0.447)	0.336 (0.264 - 0.399)	0.321 (0.250 - 0.384)
Delta from placebo		-0.001 (-0.004 - 0.002)	-0.017 (-0.04 - -0.001)	-0.059 (-0.12 - -0.016)	-0.104 (-0.18 - -0.039)	-0.117 (-0.20 - -0.048)

Note: Abbreviations are provided in the Abbreviation Listing.

Overall, the simulations suggest that the lower doses (2.5 and 5 mg QD) would not be expected to result in substantial differences relative to placebo. The highest dosing regimen (40 mg QD) is expected to result in a higher likelihood of achieving sputum NE BLQ post-baseline. However, the consequent decrease in the incidence of pulmonary exacerbations at the 40 mg QD dose is minimal when compared to that for the 25 mg QD regimen, suggesting that the value of increasing the clinical dose above that studied in INS1007-201 is limited.

Report Number: ICPD 00694-1, 2024.

The initial plan for the exposure response (E-R) analyses was to utilize the pooled data from the Phase 2 and Phase 3 studies in patients with NFCBE, Studies INS1007-201 and INS1007-301. However, given the distinct difference in sample size and duration of treatment in INS1007-301 study, the primary approach for the E-R analyses for efficacy was changed to focus on the data from the Phase 3 study.

For INS1007-301, only those subjects receiving brensocatib who were included in the population PK analysis were available for inclusion in the E-R analysis population, and all subjects receiving placebo were included.

Three clinical efficacy outcome variables from Study INS1007-301 were used as the basis for the E-R analyses for efficacy:

1) annualized rate of pulmonary exacerbations,

2) time to first pulmonary exacerbation using the postbronchodilator measurements, and

3) absolute change from baseline in postbronchodilator forced expiratory volume in 1 second (FEV1).

Safety outcome variables were confined to the adverse event of special interest (AESI) of hyperkeratosis, periodontal disease, and pneumonia. The evaluation of the impact of brensocatib exposure on the occurrence of hyperkeratosis was limited to the data from the Phase 3 study. Pooled Phase 2 and 3 data were used for the evaluation of periodontal disease and pneumonia.

The primary PK exposures, which served as independent variables in the E-R analyses, were steady-state AUC24, Cmax, and Cmin. If potential E-R relationships were identified in the exploratory analyses, statistical significance of the relationships was pursued. The statistical models used varied based upon the outcome and were matched to the Sponsor approach to the clinical data to the fullest extent possible.

The final population PK model was then used to predict brensocatib exposure in simulated patients for brensocatib doses of 10 and 25 mg once daily (QD). A similar process was also followed to predict brensocatib exposure in a hypothetical population of adolescents using age-appropriate body weight for patients aged 12 to less than 18 years.

A total of 1,189 patients from the ITT population in INS1007-301 were available for the E-R analyses (termed the E-R population); 626 patients who received brensocatib and had estimates of exposure from the population PK analysis (303 received 10 mg daily, 323 received 25 mg daily) and 563 patients who received placebo.

Ultimately, E-R relationships were identified for all three efficacy outcomes and for the one AESI that was more common in patients randomized to brensocatib (mildmoderate hyperkeratosis).

Relationships between brensocatib AUC24 and outcome were not statistically significant when AUC24 was evaluated as a continuous variable for annualized rate of pulmonary exacerbations and time to first pulmonary exacerbation.

The relationship between AUC24 and change from baseline in post-bronchodilator FEV1 at Week 52 was statistically significant when including placebo but statistical insignificant when excluding patients randomized to placebo.

Statistically significant differences in each of the efficacy outcomes were seen when patients were categorized based on threshold AUC24 values. For annualized rate of pulmonary exacerbations and time to first pulmonary exacerbation, the same threshold AUC24 of 1100 ng·h/mL was found to be discriminatory when fitting the negative binomial generalized estimating equations (GEE) model and Cox proportional hazards model, respectively.

Efficacy analysis

Annualized Rate of Pulmonary Exacerbations

The evaluation of the relationship between annualized rate of pulmonary exacerbations and brensocatib exposure was conducted primarily through the use of the negative binomial model implemented using GEE. Brensocatib exposure was evaluated as a continuous variable, using the log10-transformed AUC24, and categorically using quartiles. There appeared to be a trend for a lower annualized rate with increasing AUC24 when evaluated continuously. Patients with exposure in the third and fourth quartiles showed trends for improved outcome (rate ratio values of 0.918 and 0.868, respectively) but statistical significance was not reached.

Potential threshold AUC24 values were tested by applying the negative binomial model (without GEE) to successive datasets in which the AUC24 threshold was iteratively set at values ranging from the 5th to the 95th percentile of the AUC24 values in 10-unit intervals.

Threshold AUC24 value of 1100 ng·h/mL resulted in substantial improvement in the fit of the negative binomial model, suggesting that categorizing patients based on this threshold would be discriminatory in terms of predicting the annualized rate of pulmonary exacerbations. A total of 93 of the 303 patients (30.7%) in the E-R population who received brensocatib 10 mg had an AUC24 value below 1100. None of the patients in the E-R population who received brensocatib 25 mg had an AUC24 value below 1100.

Patients on inhaled corticosteroids at baseline have a higher annualized rate of pulmonary exacerbations. After including this variable into the model with AUC24 threshold and the stratification factors, the estimated rate ratio for patients with AUC24 above 1100 is 0.857 with a p-value of 0.050.

Time to First Pulmonary Exacerbation

While the benefit is clear in patients randomized to active drug, there is not a consistent relationship between AUC24 and the time to first pulmonary exacerbation when evaluated univariably. This finding was confirmed using a multivariable Cox proportional hazards model.

Similar to that which was seen with annualized rate, there are trends for patients in the third and fourth quartile to have improved outcome (hazard ratio values of 0.850 and 0.878, respectively) but statistical significance was not reached.

Potential threshold AUC24 values were tested in this case by applying the Cox proportional hazards model. A threshold AUC24 value of 1100 ng·h/mL was also found to be most predictive for the time to first pulmonary exacerbation outcome.

Patients with AUC24 above 1100 have a lower risk of developing a first pulmonary exacerbation at a given time after controlling for all stratification factors and significant covariate effects.

Table 53: Multivariable Cox proportional hazards model fit to the time to first pulmonary exacerbation data from the E-R population using AUC24 threshold as the measure of brensocatib exposure

Term	Estimate (SE)	Hazard Ratio (95% CI)	p-value
Significant predictors			
AUC_{24} < 1100[a]	0.200 (0.142)	1.22 (0.924 - 1.61)	0.16
AUC_{24} > 1100[a]	-0.211 (0.0834)	0.810 (0.688 - 0.954)	0.011
Inhaled corticosteroid use at baseline	0.226 (0.0846)	1.25 (1.06 - 1.48)	0.008
Race: American Indian or Alaska Native[b]	-1.36 (0.582)	0.257 (0.0823 - 0.805)	0.020
Stratification Variables			
Positive sputum sample for *Pseudomonas aeruginosa* at baseline	0.171 (0.0827)	1.19 (1.01 - 1.39)	0.039
≥3 prior PEs in previous 12 months	0.482 (0.0823)	1.62 (1.38 - 1.90)	<0.001
Region[b]			
Japan	-0.647 (0.200)	0.524 (0.354 - 0.776)	0.001
North America	0.0578 (0.114)	1.06 (0.848 - 1.32)	0.612
Rest of World	-0.193 (0.0917)	0.825 (0.689 - 0.987)	0.035
Age group – Adults[c]	0.287 (0.249)	1.33 (0.819 - 2.17)	0.248

Note: Abbreviations are provided in the Abbreviation Listing.
a. Reference is placebo.
b. Reference region is Europe
c. Reference age group is adolescents

Change from Baseline in Post-Bronchodilator FEV1

The evaluation of change from baseline in postbronchodilator FEV1 was shifted from maximum change to the change over time with primary focus on the change at the end of therapy (i.e., Week 52).

When evaluated univariably there was a trend for the lower loss in lung function, as measured by change from baseline in FEV1, with increasing brensocatib exposure. This process identified a threshold value of 1531 ng·h/mL. A total of 167 of the 301 patients (55.5%) in the FEV1 E-R population who received brensocatib 10 mg had an AUC24 value below 1531 ng·h/mL; only 2 of 312 patients (0.641%) in the FEV1 E-R population who received brensocatib 25 mg had an AUC24 below 1531 ng·h/mL.

Patients with AUC24 above 1531 ng·h/mL had a statistically significantly lower drop in FEV1 at Week 52, when evaluated univariably (p = 0.010). However, the difference between the patients above and below the threshold was not statistically significant (p = 0.148).

When compared to placebo, only brensocatib-treated patients with AUC24 above 1531 ng·h/mL had a change from baseline in FEV1 at Week 52 that was statistically significantly different from placebo (p = 0.004); the p-value for brensocatib treated patients with AUC24 below 1531, compared to placebo, was 0.983.

Safety analysis

Differences in the exposure distributions are not apparent between patients who experienced an AESI and those who did not.

Examination of the incidence of AESI by AUC quartile (Table 54) indicates no differences in incidence by quartile for periodontal disease or pneumonia. However, the incidence of hyperkeratosis in patients from INS1007-301 (mild and moderate only, no patients experienced severe hyperkeratosis) is slightly higher

in treated patients and the highest incidence (5.84%) is observed in patients in Quartile 4, suggesting that there may be a relationship between brensocatib exposure and the incidence of hyperkeratosis.

It is important to note that the point estimate for the odds ratio (3.77) indicates that the odds of developing hyperkeratosis increases by 3.77-fold with every 10-fold increase in AUC24. Given that the odds are very low for placebo (0.0003990), the expected probability at the highest predicted AUC24 of 13,883 ng·h/mL is 6.84%.

Table 54: Incidence of AESI by steady-state AUC24 quartile

AESI	AUC_{24} quartile	No	Yes
Hyperkeratosis[a]	Placebo	559 (99.3%)	4 (0.710%)
	Q1	155 (98.1%)	3 (1.90%)
	Q2	155 (98.7%)	2 (1.27%)
	Q3	154 (98.1%)	3 (1.91%)
	Q4	145 (94.2%)	9 (5.84%)
Periodontal disease	Placebo	630 (97.2%)	18 (2.78%)
	Q1	189 (96.4%)	7 (3.57%)
	Q2	186 (94.9%)	10 (5.10%)
	Q3	190 (96.9%)	6 (3.06%)
	Q4	188 (96.4%)	7 (3.59%)
Pneumonia	Placebo	611 (94.3%)	37 (5.71%)
	Q1	190 (96.9%)	6 (3.06%)
	Q2	190 (96.9%)	6 (3.06%)
	Q3	187 (95.4%)	9 (4.59%)
	Q4	181 (92.8%)	14 (7.18%)

Note: Abbreviations are provided in the Abbreviation Listing.
a. Limited to data from INS1007-301

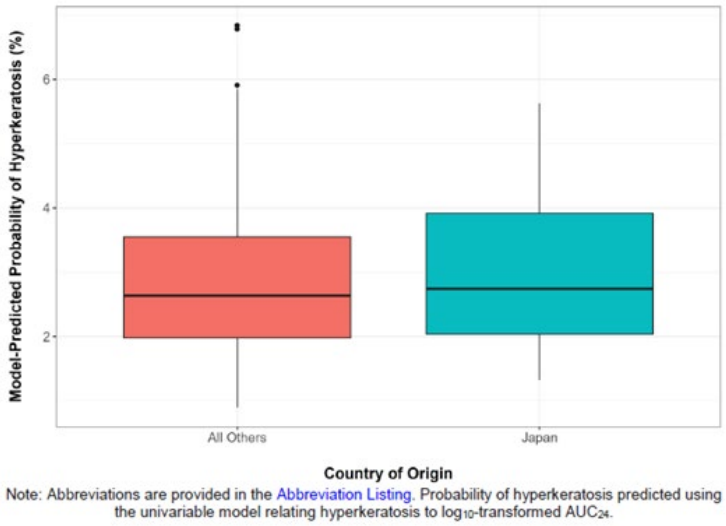
Table 55: Results of final multivariable logistic regression model fit to the hyperkeratosis data from the E-R population using log10-transformed AUC24 as the measure of brensocatib exposure

Term	Estimate (SE)	Odds Ratio (95% CI)	p-value
Intercept	-7.85 (1.45)	0.000390 (0.0000168 - 0.00532)	<0.001
log_{10} AUC_{24}[a]	1.33 (0.425)	3.77 (1.72 - 9.31)	0.002
Baseline eosinophils[b]	-2.50 (1.94)	0.0824 (0.00107 - 1.86)	0.199[c]
Asthma	0.742 (0.473)	2.10 (0.783 - 5.16)	0.117[c]

Note: Abbreviations are provided in the Abbreviation Listing.
a. Continuous variable including placebo patients, who were assigned a steady-state log_{10}AUC of 2.25 (chosen by convention of using log of 0.5 times the minimum AUC in PK patients but it also is essentially optimal by AIC).
b. Baseline eosinophils as a continuous covariate in units of $1/mm^3$. Using the categorical version (<300/L vs ≥300/L) results in an increase in the AIC.
c. Note that inclusion of these two covariates results in a decrease in the AIC despite the lack of statistical significance of the individual effects. Removal of these two covariates does not substantially impact the parameter estimate for AUC.

Given the higher exposure seen in patients from Japan, the risk of hyperkeratosis was evaluated using individual post-hoc estimates of AUC24 from the population PK analysis. The distributions of the probability of hyperkeratosis are highly concordant between the two groups with the exception of the 75th percentile, which is numerically higher in patients from Japan at 3.92% vs 3.55%).

Figure 24: Predicted probability of hyperkeratosis based on post-hoc estimates of AUC24, stratified by country of origin.



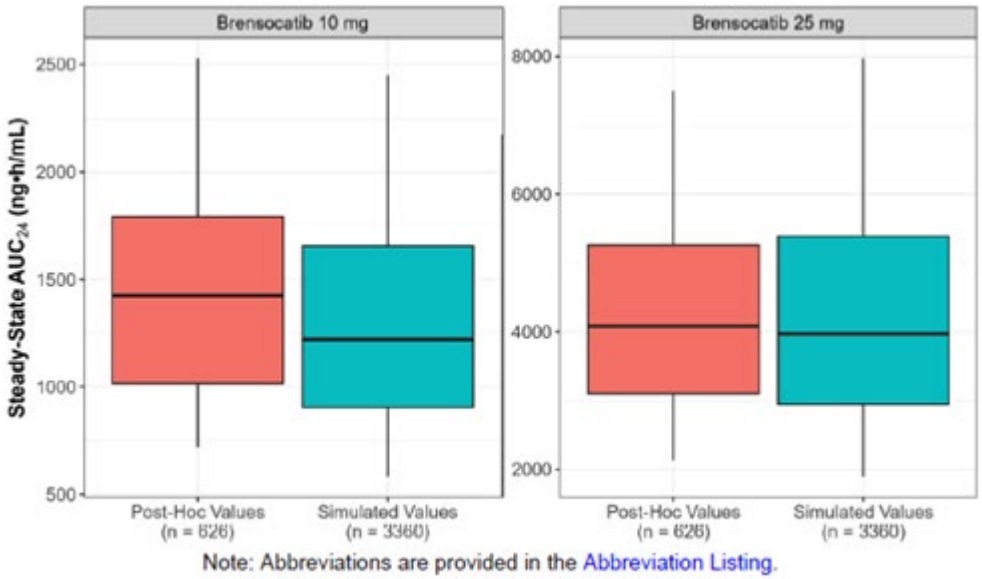
Note: Abbreviations are provided in the Abbreviation Listing. Probability of hyperkeratosis predicted using the univariable model relating hyperkeratosis to $\log_{10}$-transformed AUC_{24}.

Model-Based Simulations

Brensocatib Pharmacokinetic Exposure Predictions by Dose

The simulated exposures are slightly lower than the post-hoc estimates from patients included in the population PK analysis. On a median basis the difference is 11.4% for the 10 mg group and 4.2% for the 25 mg group. The percentage of simulated patients receiving the 10 mg dose who have AUC24 below the two thresholds are 11-14% higher than the corresponding percentages in the patients from the population PK analysis population.

Figure 25: Box-and-whisker plots showing the distributions of simulated AUC24 values in the hypothetical adult patient population and post-hoc estimates from adults from INS1007-301, by treatment group



Note: Abbreviations are provided in the Abbreviation Listing.

Annualized Rate of Pulmonary Exacerbations

While the distributions of the annualized rate overlap substantially, the trend for lower rates in patients receiving brensocatib 25 mg is apparent.

Table 56: Summary statistics of the predicted annualized rate of pulmonary exacerbations (number/yr) in the hypothetical adult patient population, by treatment group

Treatment group	Geometric mean (CV%)	Median (5^{th} - 95^{th})
Placebo	1.30 (31.1)	1.23 (0.951 - 2.27)
Brensocatib 10 mg	1.22 (33.8)	1.17 (0.801 - 2.36)
Brensocatib 25 mg	1.09 (31.2)	1.03 (0.801 - 1.91)

Note: Abbreviations are provided in the Abbreviation Listing.

Time to First Pulmonary Exacerbation

Kaplan-Meier curves show an advantage for the 25 mg dose in terms of time to first pulmonary exacerbation. The fact that the 10 mg dose is only slightly better than placebo is reflective of two factors:

1. The lower predicted AUC24 estimates relative to the post-hoc estimates from the enrolled patients, which results in 40% of hypothetical patients failing to achieve the threshold of 1100 ng·h/mL, and

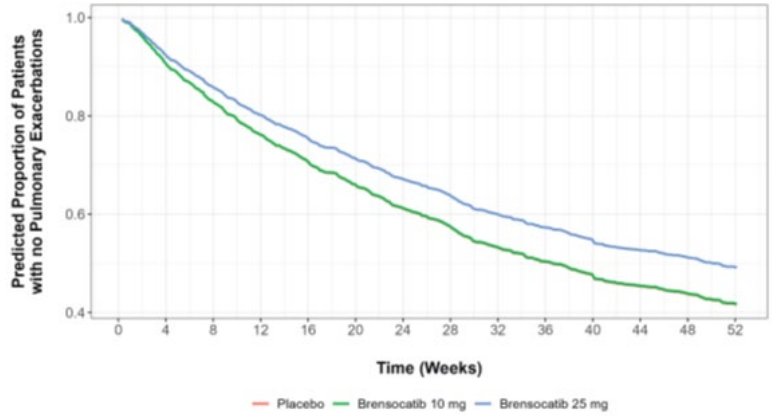
2. The use of an exposure threshold for this E-R relationship, as opposed to exposure as a continuous variable, results in an "all or nothing" behavior to the simulations such that patients with exposure below the threshold have a higher risk of a pulmonary exacerbation at a given time relative to placebo (hazard ratio of 1.22,).

Table 57: Summary statistics of relevant output from the simulated time to first pulmonary exacerbation output, stratified by treatment group

Treatment Group	Probability of being free of pulmonary exacerbations		Time to first pulmonary exacerbation (weeks)[a]
	Through 6 months	Through 1 year	
Placebo	0.597 (0.404 - 0.707)	0.417 (0.215 - 0.555)	37 (29 - 50)
Brensocatib 10 mg	0.598 (0.360 - 0.755)	0.418 (0.177 - 0.621)	37 (27 - >52)
Brensocatib 25 mg	0.658 (0.480 - 0.755)	0.492 (0.288 - 0.621)	51 (38 - >52)

Note: Abbreviations are provided in the Abbreviation Listing.
Note: Summary statistics presented as median (5^{th} – 95^{th}).
a. These summary statistics represent the median (5^{th} – 95^{th}) of the median values for each hypothetical patient.

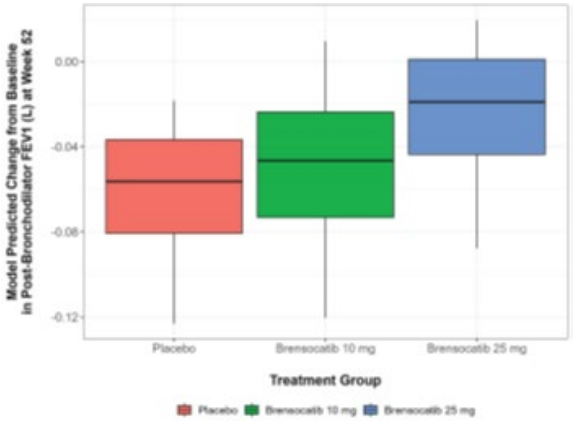
Figure 26: Predicted time to first pulmonary exacerbation in the hypothetical adult patient population, by treatment group



Change from Baseline in Post-Bronchodilator FEV1

Overall, the predictions from the linear, mixed-effects model for change from baseline in post-bronchodilator FEV1 show a trend for less loss in lung function, as measured by change from baseline in post-bronchodilator FEV1, for patients receiving brensocatib 25 mg.

Figure 27: Box-and-whisker plots showing the distributions of change from baseline in post-bronchodilator FEV1 at Week 52 in the hypothetical adult patient population, by treatment group



Adverse Events of Special Interest (Hyperkeratosis)

As expected, given the results of the E-R analysis for hyperkeratosis, the risk of hyperkeratosis is expected to be higher at the 25 mg dose. However, the predicted probability is low (median [5th – 95th] of 3.15% [1.15% – 7.17%] at the 25 mg dose).

Table 58: Summary statistics of the predicted probability of hyperkeratosis (%) in the hypothetical adult patient population, by treatment group

Treatment group	Geometric mean (CV%)	Median (5th - 95th)
Placebo	0.520 (62.9)	0.556 (0.216 - 1.25)
Brensocatib 10 mg	1.55 (67.0)	1.62 (0.587 - 3.77)
Brensocatib 25 mg	3.01 (66.3)	3.15 (1.15 - 7.17)

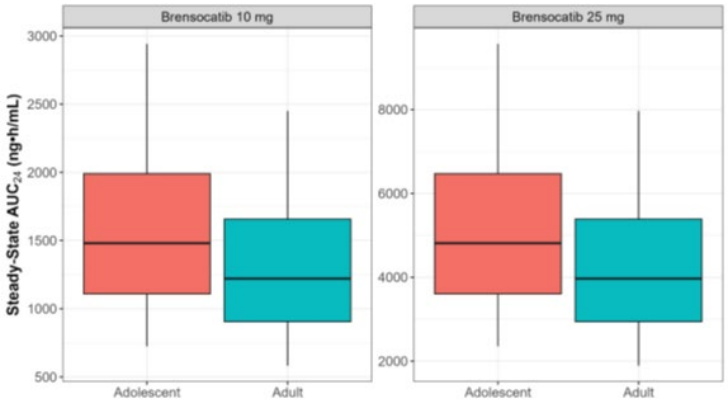
Note: Abbreviations are provided in the Abbreviation Listing.

Model-Based Simulations for Adolescents

Brensocatib exposures were simulated for a hypothetical population of adolescents with NCFBE by replacing the observed age in the patients from the hypothetical adult population with a random age between 12 and 18 years and then simulating an age-appropriate body weight based on the simulated age and observed sex.

The distribution of weights in the hypothetical adolescent population was lower than that in adults. The impact of this lower weight on predicted AUC24 is shown in figure below which show that the predicted exposure was higher in the hypothetical adolescent patient population.

Figure 28: Box-and-whisker plots showing the distributions of simulated AUC24 values in the hypothetical adolescent and adult patient populations, by treatment group



As expected, the probability was higher for adolescents, regardless of dose, but the overall probability was only slightly higher than in adults and remains low (median [5th – 95th] of 3.50% [1.32% – 8.03%] at the 25 mg dose).

Figure 29: Box-and-whisker plots showing the distributions of probability of hyperkeratosis in the hypothetical adult patient population, by treatment group

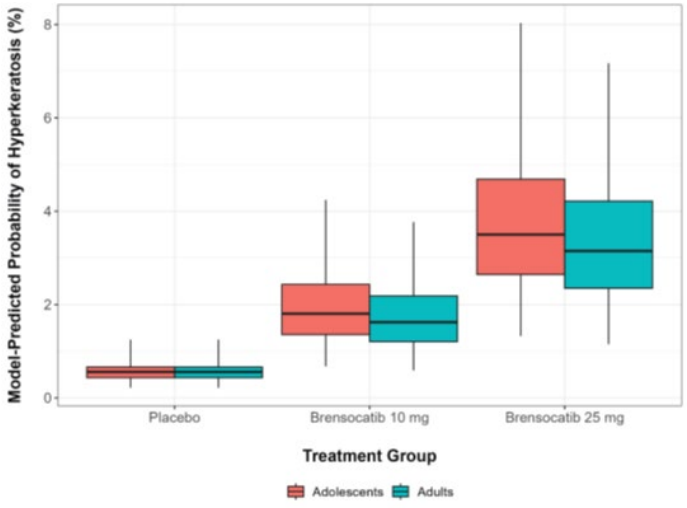


Table 59: Summary statistics of the predicted probability of hyperkeratosis (%) in the hypothetical adolescent patient population, by treatment group

Treatment group	Geometric mean (CV%)	Median (5th - 95th)
Placebo	0.520 (62.9)	0.556 (0.216 - 1.25)
Brensocatib 10 mg	1.73 (66.8)	1.81 (0.675 - 4.24)
Brensocatib 25 mg	3.36 (66.0)	3.50 (1.32 - 8.03)

Note: Abbreviations are provided in the Abbreviation Listing.

6.2.5. Dose selection and therapeutic window

Prior to advancement to Phase 2, Phase 1 single dose and MAD studies in healthy volunteers were conducted with single dose administration at 5 to 65 mg and repeated dosing at 10 to 40 mg QD. The

clinical PK and PD data indicated that brensocatib provided adequate steady-state exposures at the proposed doses of 10 and 25 mg QD for further evaluation in the Phase 2 study in a population with NCFBE. The results of Phase 1 multiple ascending doses demonstrated further that plasma NE activity was reduced with increasing doses asymptotically approaching a flat relationship above 25 mg.

Based on the plasma NE activity and safety profile, 10 and 25 mg QD were selected to be tested in the Phase 2 Study INS1007-201.

In the final PK/PD model, which incorporated data from the Phase 3 clinical study, the 25 mg dose was found to be more clinically relevant compared to the 10 mg dose, which demonstrated only marginal improvement over placebo with respect to the relevant efficacy endpoints. This is in contrast with the in vivo results from the Phase 3 clinical study, where the 10 mg dose showed greater clinical relevance.

Assessment of relevant covariates in the clinical studies as well as in the population PK (popPK) model did not identify any significant changes in brensocatib exposure that would necessitate dose adjustment. However, when covariates were assessed in combination, the model revealed a significant change in brensocatib exposure.

Systemic exposure in adolescents was observed to be somewhat higher than in adults, which could potentially increase the risk of adverse effects in the adolescent population. Nevertheless, the probability of adverse event occurrence is similar between adolescents and adults.

6.2.6. Overall discussion and conclusions on clinical pharmacology

6.2.6.1. Discussion

Primary pharmacology

Brensocatib is introduced as the first-in-class small-molecule, oral, competitive and reversible inhibitor of DPP1. DPP1 activates pro-inflammatory NSPs during neutrophil maturation in the bone marrow. Activated NSPs are implicated in the pathogenesis of many neutrophil-mediated inflammatory diseases, including bronchiectasis. Brensocatib reduces the activity of NSPs including NE, CatG, and PR3. PD measurements of NSP activity in healthy volunteers (Study D6190C00001) revealed a maximum reduction in NE activity about 7 to 10 days after the last dose of brensocatib with a dose-dependent effect across the 10, 25, and 40 mg dose levels. In the target population (studies 201, 211, and 301), brensocatib reduced NSP activity in blood and sputum. In NCFBE patients recruited in phase 3 Study INS1007-301, the time to maximum reduction in NSP activity was observed about 4 weeks after treatment initiation. Again, the effect was dose dependent (10 mg, 25 mg brensocatib vs placebo), and NSP activity recovered to normal within the 4-week follow-up. Across studies, variability was observed within and between individuals and studies. While DPP1 inhibition itself was not directly measured, NSP suppression across different compartments supports the intended mechanism. Given the consistent efficacy signals, limitations in PD assessment (e.g., variability, missing DPP1 data) are not considered critical.

<u>Bioanalytical methods</u>

Plasma samples were processed using protein precipitation followed by liquid chromatography coupled with tandem mass spectrometry (LC-MS/MS), whereas urine samples were prepared by dilution prior to LC-MS/MS analysis. The validation data demonstrated that all methods met the predefined acceptance criteria for accuracy, precision, selectivity, sensitivity, linearity, and stability.

Cross-validation of bioanalytical methods used across different studies was performed and yielded satisfactory results, supporting the consistency and reliability of the analytical approaches. Incurred

Sample Reanalysis (ISR) results were reported to be within acceptable limits, indicating reproducibility of the assay performance in study samples. The validated methods are considered fit for purpose, and the resulting pharmacokinetic data are deemed reliable and suitable for the clinical evaluation of brensocatib.

In addition, the applicant developed and validated methods for the quantification of thiocyanate, verapamil, itraconazole, hydroxyitraconazole, and moxifloxacin and pharmacodynamic biomarkers (NE, PR3, and CatG). The validation of these methods was considered adequate, and the analytical performance is deemed acceptable.

Formulations used during clinical development

A comprehensive PK/PD programme was presented that was also guided by SA procedures during the PRIME regulatory support process. Three different formulations were applied during clinical development, comprising an oral solution, used during early SAD/MAD Study D6190C00001 and DDI Study D6190C00003, and phase 2 and phase 3 film-coated tablet formulations. The intended commercial formulation of 25 mg is identical for the core tablet formulation, shape, and size used across the clinical development programme, except for changes in the film coating colour from brown used in the clinical studies to grey for the commercial drug product. Considerable efforts were undertaken to demonstrate *in vitro* dissolution similarity across the tablet formulations. Comparability between phase 2 and phase 3 tablets was considered acceptable based on further justification and dissolution results at different pH provided by the applicant.

The phase 2 tablet formulation has only been used in one study (INS1007-101) comparing brensocatib BA after SD and MD in Japanese vs Caucasian subjects. Pop PK modelling revealed BE between the phase 2 and phase 3 tablet formulations.

Evaluation and qualification of models

Population PK models

The initial popPK model was based on steady-state data from studies INS1007-101 (healthy volunteers, n=68) and INS1007-201 (NCFBE patients, n=158), as pooling with both the single-dose and steady-state data failed due to model convergence issues. The final dataset included 225 subjects and 1,284 concentrations. Absorption and distribution were explored through various models, though details were not provided. Scaling of CL/F, Vc/F, and Vp/F to body weight was necessary for model convergence.

The final model was a two-compartment model with linear elimination and six transit compartments, using separate Ka values for fed and fasted states. Age (on Vc/F) and renal function (on CL/F) were statistically significant covariates but not clinically relevant. The model showed good fit, though overprediction occurred for the non-relevant 10 mg dose group.

A second popPK model was developed using pooled data from 11 studies (Phase 1–3), comprising 1,098 subjects and 11,643 concentrations. The final popPK model, using all data available, was best described by two systemic compartments with linear elimination and an oral depot compartment with an associated dose-dependent relative bioavailability, a 3-compartment transit chain, and first-order absorption described by separate Ka values for fed and fasted states for description of the absorption profile. Parameters were precisely estimated with good GOF and VPC plots.

Covariates affecting variability included formulation, CYP3A4-modifying drugs, hepatic function, race, and weight. While statistically significant, their individual impact was not clinically relevant, though combined effects may be more pronounced. Despite some limitations, the model is considered adequate for describing the data.

PBPK models

The applicant developed a PBPK model to waive clinical DDI studies and assess brensocatib as both a CYP3A victim and perpetrator in healthy subjects. Additionally, simulations evaluated the effect of gastric pH and brensocatib's impact on midazolam (CYP3A substrate) and metformin (MATE substrate).

The model, based on *in vitro* and *in vivo* data (mass balance, SAD/MAD, and verapamil DDI studies), showed good agreement with observed Cmax for a 35 mg dose but overpredicted Cmax by 1.25-fold in the verapamil study. Further verification using data from study D6190C00001 and a DDI study with itraconazole revealed the following:

- Overprediction of Cmax >1.25-fold for a 5 mg dose.
- Acceptable prediction for higher doses, though underprediction increased with dose.
- In multiple-dose studies, AUC was overpredicted at lower doses but improved at steady state.
- Significant overprediction (Cmax 2.23-fold) in the itraconazole DDI study.

Simulations indicated that rifampicin and efavirenz reduced brensocatib exposure by ~50% and 30%, respectively, without affecting Cmax. Gastric pH changes had no impact on brensocatib exposure or Cmax. As a perpetrator, brensocatib did not alter midazolam or metformin exposure or Cmax at 65 mg.

Subsequent clinical DDI studies (Study 106 with rifampicin and esomeprazole, and Study 109 with clarithromycin) reduced the regulatory reliance on the PBPK model. The model failed to predict metformin interaction despite *in vitro* MATE inhibition data and did not include other transporters (e.g., BCRP, OATP1B1).

Bioequivalence between oral solution and tablet formulations was not established based on this model. Model overprediction after single 10 mg and 25 mg doses using the oral solution suggests formulation-specific absorption differences. The itraconazole DDI study, also using the solution, showed poor prediction accuracy (Cmax and AUC both above acceptable cutoffs).

There is uncertainty in the model inputs: ka, fu, and Papp values were inconsistent with other datasets, and *in vitro* CYP3A4 inhibition data did not align with *in vivo* exposures.

Due to data limitations, formulation differences, and poor model performance in key scenarios, the PBPK model is not considered suitable for further simulation or regulatory decision-making However since PBPK model is not needed for further assessment of DDI given that there is siffucient data from *in-vitro* and *in-vivo*, there is no need for update of PBPK model.

Pharmacokinetics

Absorption

Absorption was described as fast, with Tmax being approximately 1-hour post-dose in all clinical studies. In the final PopPK model, an oral depot compartment with an associated dose-dependent relative bioavailability, a 3-compartment transit chain, and first-order absorption described by separate Ka values for fed and fasted states were used to describe the absorption profile.

Bioequivalence

Three formulations were used during clinical development: an oral solution (used in studies D6190C0001 and D6190C0003) and two tablet formulations for Phase 2 and Phase 3. While the tablet core remained unchanged between Phase 2 and 3, the outer coating differed.

To support formulation waivers, two popPK models were developed: one comparing the oral solution and Phase 2 tablet, and another comparing Phase 2 and Phase 3 tablets. The oral solution showed faster absorption and slightly higher exposure than tablets, which was addressed in the model by adjusting the absorption rate constant.

For the oral solution vs. tablet comparison, the popPK model (report 00566-1) was developed using data from studies 101 and 201. Although initially pooled with data from four studies, only Phase 1 data were used in the final model. Despite model-based simulations showing bioequivalence at steady state (40 mg dose), this dose is not clinically relevant, and steady state may mask absorption differences. Per guidelines, single-dose comparisons are preferred for detecting formulation differences. Thus, the claim of bioequivalence between oral solution and tablet is weak. However, similar SAD, MAD, and food-effect data were collected for both formulations, supporting comparability to some extent. Furthermore, the applicant performed clinical study 110 with aim to assess relative bioequivalence between oral solution and Phase 3 tablet formulation after single dose administration of 25 mg brensocatib. Results of the study indicate bioequivalence between oral solution and Phase 3 tablet formulation.

For the Phase 2 vs. Phase 3 tablets, a modified popPK model was applied, incorporating transit compartments and fewer structural compartments. Two approaches were used:

1. Direct approach: Post-hoc individual PK parameters were compared using standard BE methods. Bioequivalence for AUC_{0_24} and Tmax was shown, but not for Cmax after a single dose (90% CI exceeded limits). At steady state, BE was demonstrated.

2. Model-based simulation: 100 virtual BE studies (n=16 each, 25 mg dose, crossover design) showed BE for both Cmax and AUC within accepted 90% CI limits.

Therefore, while the Phase 2 and 3 tablet formulations may be considered bioequivalent based on simulations, the oral solution vs. tablet comparison lacks robust support, particularly due to the use of non-clinical doses and reliance on steady-state data.

Distribution

In vitro studies suggest moderate protein binding, from 82.2% to 87.2%. Fraction unbound in healthy volunteers in the study assessing hepatic impairment was around 17.8%. Mass balance study showed that the whole blood to plasma ratio is 0.716, suggesting insignificant binding to blood cells.

Volume of distribution in the final popPK model was estimated to be Vc/F (L/70 kg) 78.8, and for the peripheral compartment (Vp/F) 50.4 L. The same model was used to predict volume of distribution for clinical studies with target population (301 and 201). This indicates that volume of distribution is much lower than in healthy volunteers.

After oral administration, the volume of distribution at steady state was 126-138 L (CV: 22.4-23.3%) in adult patients and 71.3-83.6 L (CV: 19.9-26.3%) in adolescents with NCFB. The SmPC information accepted in section 5.2 is based on popPK model and is considered acceptable.

Elimination/Metabolism

In the final pharmacokinetic (popPK) model, the estimated clearance (CL) of brensocatib in the target population was between 2.81–3.02 L/h in adults and 2.07–2.3 L/h in adolescents. The estimated terminal half-life (t1/2) was 32.6-39.6 hours (CV: 26.6-33.0%) in adult patients and 26.9-27.8 hours (CV: 26.8-37.3%) in adolescent patients.

These findings indicate that clearance in the target population is approximately 50% lower, and the half-life is moderately prolonged compared to values observed in healthy volunteers.

<u>Metabolism</u>

In HV (mass balance study 103), brensocatib was moderately metabolized primarily via oxidation, hydrolysis, oxidative dealkylation, sulfuration, and carbamoyl glucuronidation. The overall mean recovery of radioactivity in urine and faeces over the 312-hour sampling period was 82.5%, with individual subject recoveries ranging from 80.0% to 86.3%. Across all excreta, approximately 27

metabolites were detected, of which only 9 were structurally identified. Following a single oral dose of radiolabelled brensocatib, 54.2% of dose was excreted in urine and 28.3% in faeces with most radioactivity excreted within 72 hours.

The major component present in excreta was the unchanged parent compound (brensocatib). Among the 27 quantifiable metabolites in plasma, urine, and feces, none of the metabolites in urine and feces was considered major (each <10% dose). Six of the metabolites were detected in plasma including 5 minor metabolites (each <0.5% of total AUC) and 1 major metabolite, M8 (thiocyanate, an endogenous compound).

The issue of metabolite M8 characterization was previously discussed during EMA/SA/0000126545. In mass-balance study 103, the thiocyanate (M8) metabolite was the most abundant component in plasma (51%), but a trace urinary component. Thiocyanate is endogenous in human plasma and cells. Plasma levels of thiocyanate were taken during phase 2 study 201 in NCFBE patients. Both for brensocatib and placebo the concentration of thiocyanate remains well below levels that are provided as normal. Baseline corrected thiocyanate levels did not rise during QD administration of brensocatib 25 mg over 200 days.

With respect to the mass balance study, it is critical that the positioning of the radioactive label (^{14}C) ensures its retention in all relevant metabolites, including those resulting from minor structural modifications. In this study, the radiolabel was positioned in the nitrile moiety of the brensocatib molecule, which is susceptible to metabolic cleavage. As a result, any metabolites formed through decyano metabolic pathways would not retain the radiolabel and, therefore, remain undetected and uncharacterised in the current study design. However, the process of M8 formation is slow, M8 was first detected at 24 hours post-dose, i.e. the metabolic stability of radiolabeled brensocatib is considered adequate.

Concerns have arisen from the observation that in plasma, M8 was the only major metabolite, corresponding to 51% of total plasma radioactivity AUC. It was elucidated, however, that this is mainly due to M8's extremely long half-life (7-8 days). All other metabolites identified were minor (individual AUC < 1% of total plasma radioactivity AUC), and were unquantifiable at 24 hours post-dose as well as at later time points. Besides M8, no other late forming metabolites were detected in plasma.

The metabolic profile of brensocatib in plasma, urine, and feces was characterized over 13 days (312 hours sampling). Like in plasma, in urine and feces, there were no late forming metabolites detected based on the data from pooled urine samples (0-240, 48-96, 120-168 and 192-312 hours) and feces samples (0-216, 48-96, 120-168 and 192-312 hours), and all quantifiable metabolites were present at 48-96 hour in urine and/or feces samples.

M8 was first detected 24 hours post-dose, and the overall amount of M8 formed was estimated to be <5% (0.11% in early pooled urine sampling [0-240 h], 0.806% in last sample [192-312 h], plus 3.49% estimated unrecovered [<312 h]). The prerequisite for metabolites leaving undetected is the cleavage of the radiolabeled nitrile side chain (the M8 metabolite). Given the small overall amount of M8 (<5% of radioactive dose in urine), the potential for leaving late-forming (potentially major) metabolites undetected, is considered very small. Mass balance study 103 is considered sufficient to characterize the metabolic profile of brensocatib.

PK results

In first-in-human SAD Part 1a of Study D6190C0001 ascending single doses of brensocatib were administered across the 5 mg to 65 mg range using an aqueous solution. Across the full dose range, brensocatib was rapidly absorbed with Tmax values of 0.5 to 0.75 h. The extent (AUC) and rate of exposure (Cmax) increased with dose in a larger than dose proportional manner (dose proportionality slope estimate about 1.3 for both parameters). Terminal elimination half lives of brensocatib were

around 20-25 hours. The fraction of the dose excreted in the urine as unchanged drug was moderate for all dose levels, with 11.1%, 13.5%, 19.9%, 16.5% and 18.0% being eliminated up to 48 hours post-dose at the 5, 15, 35, 50 and 65 mg single dose levels, respectively.

During the MAD Part 2 of Study D6190C0001 three dose levels of brensocatib (10, 12, 40 mg) were tested for once daily multiple dosing for up to 28 days. Following multiple once daily administration at 10 mg, 25 mg, and 40 mg, the steady state Cmax and AUCτ increased in an approximately dose proportional manner. There was 2.3- to 2.5- and 1.7- to 1.9-fold accumulation of brensocatib following multiple dosing, compared to the data from the single dose administration on Day 1 based on AUCτ and Cmax, respectively.

Given the observed elimination t1/2 of 24-34 h in MD studies D6190C0001 and INS1007-101, achievement of steady state would normally be expected after about 4-5 half-lives, i.e. by Day 7. Based on measured pre-dose concentrations of brensocatib, however, it is considered that stable steady state plateau levels were obtained only by about 14 days of daily dosing.

Later in clinical development, further single and multiple dose data were obtained from Study INS1007-101 for the 10 mg, 25 mg, and 40 mg dose, administered as tablets. The study also included a comparison between the Caucasian and the Japanese population and examination of the food effect. About dose proportional increases in exposure were observed across the 10-40 mg dose range. Statistical analysis of the effect of Japanese/Caucasian descent on INS1007 dose-normalized PK parameters showed that exposure was moderately greater for Caucasian subjects compared to Japanese subjects, which is not of concern within the present European MAA.

Brensocatib is time-independent as half-life did not change after single and multiple dosing.

Intra- and inter-individual variability

Based on the final popPK model, brensocatib showed low to moderate IIV, which was estimated to be the 22.2%, 15.3%, and 11.5% for Vc/F, Vp/F, and relative bioavailability (relF), respectively. The IIV in Ka and CL/F was somewhat more pronounced (40.5% and 34.7%, respectively). Similar was shown for intrasubject variability.

Food effect

After administration of a 40 mg SD, in the Caucasian population the point estimators for the fed/fasted ratio were close to 1 (Cmax 101.24, AUClast 101.31). The observation that the upper 90% CI of Cmax was slightly above the 80-125% acceptance range (Cmax 90% CI: 81.41, 125,91) is of less concern and may be explained by the small sample size (n=10). Additional data regarding a potential food effect were generated in Part 1b of Study D6190C0001 after administration of 35 mg brensocatib, using the early oral solution formulation, in n=6 subjects. Overall, the CHMP agreed that a clinically food effect was not observed and brensocatib may be taken irrespective of food as stated in the SmPC.

Target population

PK data were obtained in the NCFBE target population from phase 2 study INS1007-201 and phase 3 study INS1007-301. The PK profile of brensocatib in the target population was comparable to that obtained in healthy volunteers. Brensocatib was rapidly absorbed with Tmax around 1 hour. Across the 10 and 25 mg dose, the rate and extent of brensocatib exposure increased with dose. In study INS1007-201, plasma levels (Cmax and AUC) were dose proportional under steady state conditions. In study INS1007-301, an approximately 2-fold accumulation ratio was observed after multiple dosing for both dose levels (10 mg, 25 mg), while in study INS1007-201 accumulation showed greater variability, however, was in the same order of magnitude (drug accumulation in Cmax and AUCs was 2.5- to 4.6-fold at 10 mg and 2.2- to 3-fold at 25 mg).

In study INS1007-301, PK data were also generated in a small subset of adolescent NCFBE participants. The average adolescent post-dose concentration was approximately 34% to 36% higher than that in the adults from the substudy. Overall, plasma levels in adolescents were generally comparable to those in adult patients.

Based on the population pharmacokinetic (popPK) model, clearance was lower, half-life was moderately prolonged, and volume of distribution was reduced compared to healthy subjects. However, Cmax and AUC values were generally comparable between populations.

Special populations

The applicant assessed renal and hepatic impairment, along with demographic and clinical covariates, using clinical studies and population PK modelling. This approach is acceptable.

Renal impairment

The impact of renal impairment on brensocatib exposure was examined in Study INS1007-102 in participants with mild, moderate, and severe RI following a single oral administration of 25 mg of brensocatib as compared to healthy controls. Plasma levels of brensocatib in subjects with mild RI were overlapping with those of healthy controls. Somewhat unexpectedly, AUC values were increased by 27% in moderate RI, but decreased by 28% in severe RI, as compared to healthy volunteers. Given small sample sizes of N=6, it remains unclear whether these findings are actually related to the degree of renal impairment. Recruitment of n=6 subjects per RI category is at the lower end, however, acceptable according to EMA Guideline EMA/CHMP/83874/2014. Overall, study INS1007-102 did not point to a clear relationship between brensocatib exposure and renal impairment. Based on study INS1007-102 results, the CHMP agreed that dose adjustment for participants with mild, moderate, or severe renal impairment is not necessary as stated in the SmPC sections 4.2 and 5.2.

Hepatic impairment

In Study INS1007-105, brensocatib PK and safety were evaluated in participants with mild (Child-Pugh score 5 to 6), moderate (Child-Pugh score 7 to 9), and severe (Child-Pugh score 10 to 12) HI as compared to healthy controls following a single oral administration of 25 mg of brensocatib.

The unbound PK parameters were derived based on the percent unbound estimates determined by the plasma protein binding experiments conducted in Study 8496474. The mean fraction of unbound (funb; average of estimates based on 4- and 24-hour samples) was 16.9%, 19.7%, and 26.9% across the HI groups and 17.8% in the normal hepatic function group. This was found highly correlated to the baseline serum albumin concentration, suggesting that brensocatib is mainly bound to albumin in the circulation, and the percent unbound appeared to increase with the severity of HI.

Both for AUC and Cmax, as well as for evaluation of total and unbound brensocatib, the 90%CIs spanned 1 for comparison between normal hepatic function and subjects with mild / moderate / severe HI. Hence, study 105 did not point to a significant clinically relevant influence of hepatic function on brensocatib PK. Based on the brensocatib exposure and elimination data from study INS1007-105, the CHMP agreed that dose adjustment for participants with mild, moderate, or severe hepatic impairment is not necessary as stated in the SmPC section 4.2.

Sex

Females have ~10% lower clearance than males per final popPK model. No dose adjustment is required.

Body Weight & Adolescents

Higher exposure was predicted and observed in low-weight individuals, including adolescents. However, exposure overlapped with adult data, and therefore no dose adjustment is warranted.

Special population

Population pharmacokinetic analysis showed no evidence of a clinically significant effect of age (range: 12 to 85 years), sex, race/ethnicity or body weight (range: 32 to 155 kg) on the pharmacokinetics of brensocatib.

Interaction Studies

In vitro

In vitro study (ADME-AZS-Wave3-140531) using recombinant CYP bactosomes indicates that brensocatib is predominantly metabolized by CYP3A4/5 (>97%), with minor contributions from CYP2C8 (<2%) and CYP2D6 (<1%). However, these represent relative contributions among the 10 CYP enzymes tested. The possibility of metabolism by other enzymes not included in this assay cannot be excluded.

Despite moderate clearance observed in recombinant CYP assays, brensocatib was found to be stable in both human hepatocytes (BE000083-57) and intestinal microsomes (BJAA-0003-DV-HB). The limited metabolism observed can be attributed to high test concentrations of brensocatib in relation to its moderate clearance, as well as limited sensitivity of the assays used.

Brensocatib was tested as a reversible inhibitor of CYP1A2, 2C9, 2C19, 2D6, and 3A4/5 using human liver microsomes at concentrations up to 30 µM. The Ki value for CYP3A4/5 inhibition was estimated to be >15 µM, but true Ki could not be determined, as concentrations >30 µM were not tested. The cut-off value for intestinal CYP3A4/5 inhibition was calculated as 23.78 µM, based on EMA guidance. However, based on the available data, the CYP3A4/5 Ki value was determined to be approximately 50 µM, which exceeds the cut-off value. Although the calculated Ki is only a rough estimate, the value is considered sufficiently high not to warrant additional clinical drug–drug interaction studies.

Study ADME-AZS-Wave3-140530 tested TDI of CYP1A2, 2C9, 2C19, 2D6, and 3A4/5 at 10 and 50 µM. No TDI was observed. This result is acceptable and supported.

The applicant submitted several *in vitro* studies to assess the CYP induction potential of brensocatib. Studies PR13028/PR13074/PR13101 evaluated induction of CYP1A2, CYP2B6, and CYP3A4 at concentrations up to 30 µM. However, these studies were conducted in single-replicate format (monoplicate), used different concentrations, and employed a 1-day incubation period rather than the 3-day period recommended by the EMA Guideline. Additionally, phenobarbital was used at a concentration above the recommended maximum. These studies are provided as initial screening assays are not considered predictive of clinical CYP induction however, despite mentioned deviations, all three studies showed >2-fold CYP3A4 mRNA induction in a dose dependent manner, with variable results for CYP1A2 and CYP2B6.

Study 8492164 followed EMA guidelines and tested CYP1A2, 2B6, 2C8, 2C9, 2C19, and 3A4 over 3 days, using concentrations up to 5.232 µM. Although the overall results were inconclusive, >2-fold induction of CYP3A4 and CYP2B6 was observed in one donor (donor A). Accordingly, a potential for CYP induction by brensocatib at clinically relevant concentrations cannot be excluded. The information in SmPC section 4.5 has been revised to state that *in vivo* induction of CYP2B6 and CYP3A4 cannot be excluded. Furthermore, the applicant has committed to perform an *in vivo* CYP3A4 induction study.

The section 5.2 of the SmPC reflects the effect of brensocatib on other medicinal products as follows:

[..In vitro data and population pharmacokinetic analyses indicate that brensocatib is unlikely to inhibit or significantly induce the activity of CYP isozymes or drug transporters at clinically relevant dose levels. However, in vitro studies were inconclusive regarding the potential of brensocatib to induce CYP2B6 and CYP3A4, and in vivo induction cannot be excluded…]

Brensocatib exhibits passive permeability with a Papp of 9.03 × 10^{-6} cm/s and is a substrate of both P-glycoprotein (P-gp) and BCRP, as shown by an efflux ratio of 3.86, which was reduced in the presence of verapamil and itraconazole.

For transporter inhibition, the Cheng-Prusoff equation (Ki = IC50/(1+[S]/Km)) was used to determine Ki values from the determined IC50 values. Brensocatib did not inhibit any of the tested transporters. Although OCT1 and BSEP inhibition were not tested, this is not considered critical due to the absence of relevant risk factors: brensocatib is not an OCT1 substrate, lacks OCT1/OCT2 structural alerts or hepatotoxicity signals, and shows low hepatic and biliary exposure.

In vivo DDI Studies

In study D6190C00003, co-administration with verapamil (moderate intestinal CYP3A4 and P-gp inhibitor) increased brensocatib exposure (Cmax ↑1.53-fold; AUC ↑1.32-fold), while itraconazole (strong CYP3A4 and P-gp inhibitor) unexpectedly reduced Cmax (↓0.61-fold) and caused only a modest AUC increase (↑1.14-fold). The reduction in Cmax observed with itraconazole is attributed to to the oral solution formulation used to administer itraconazole oral solution (Sporanox®, 10 mg/mL), which contained hydroxypropyl-β-cyclodextrin (HβCD), which was demonstrated to lead to unexpected drug interactions due to the victim drug absorption being limited by the high amount of HβCD. Additionally, verapamil's dual inhibition of intestinal CYP3A4 and P-gp likely enhanced oral bioavailability of brensocatib but is not considered clinically relevant. Clarithromycin, a strong CYP3A4 and moderate P-gp inhibitor, led to the largest increase in exposure (Cmax ↑1.56-fold, AUC ↑1.68-fold), supporting an additive effect on efflux and metabolism inhibition (see Study 109 below).

To further explore the potential effect of CYP3A4 inhibition, DDI Study INS1007-109 was conducted to examine the impact of the strong CYP3A4 / P-gp inhibitor clarithromycin on brensocatib absorption. When brensocatib was administered in the presence of steady-state exposure to clarithromycin, a CYP3A4 and P-gp inhibitor, the AUCtlast, AUCinf, and Cmax were increased by 56%, 56%, and 68%, respectively, and t1/2 was delayed by approximately 4 hours. The PK data from this study demonstrated that CYP3A4 and P-gp inhibition significantly increased the systemic exposure and reduced the rate of elimination of brensocatib. It is agreed that the observed interaction does not require dose adaptations to be included in SmPC section 4.2.

In conclusion the section 4.5 related to in vivo induction is reflected as follows: In vivo induction cannot be excluded. Co-administration with CYP3A4 substrates used in bronchiectasis (e.g. inhaled corticosteroids, macrolide antibiotics or inhaled bronchodilators such as salmeterol or vilanterol) may result in decreased plasma concentrations and reduced therapeutic effect. Adjustment of the concomitant treatment may be considered if efficacy is reduced.

Given the known effect of CYP3A4 inhibition on brensocatib absorption and the established pH dependency of brensocatib solubility with decreased solubility in more alkaline media, DDI Study INS1007-106 was conducted with rifampin and esomeprazole.

In study INS1007-106, rifampin (a CYP3A4 and transporter inducer via PXR activation) reduced Cmax by 15%, AUC by 33%, shortened $t_{1/2}$, and increased CL/F, consistent with induction of CYP3A4 and possibly BCRP. These results support the involvement of CYP3A4 in the systemic clearance of brensocatib. In contrast, esomeprazole had no relevant impact on brensocatib exposure, aside from a minor Tmax delay, indicating no pH- or weak CYP3A4-related interaction. Co-administration of brensocatib with CYP3A4-inducing agents or PPIs does not warrant particular dose adaptations.

Inter-conversion

Even though brensocatib is a chiral molecule, a possible inter-conversion was not discussed by the applicant. Stability testing of drug substance and drug product indicates that the chiral centers were

stable. Furthermore, data from in-vivo mass balance study were supportive and acceptable.

Genetic polymorphism

Brensocatib has been identified as a substrate of CYP3A4/5 and efflux transporters BCRP and P-gp. No clinically relevant effects are expected from polymorphism of these enzymes and transporters.

Effects of Brensocatib on QT Interval

Brensocatib, when administered as single oral doses of 40 mg (therapeutic) and 120 mg (supratherapeutic), does not have a clinically meaningful effect on QTc interval prolongation. The observed QTc changes remained well within acceptable regulatory limits, with a maximal placebo-corrected change (ΔΔQTcF) below the 10 ms threshold of concern, even at peak plasma concentrations.

Overall, the thorough QT assessment supports the cardiac safety profile of Brensocatib at both therapeutic and supratherapeutic doses. These results align with ICH E14 guidance for negative TQT studies.

Pharmacokinetics-Pharmacodynamics (PK/PD)

Evaluation and Qualification of PK/PD Models

The applicant conducted two PK/PD modelling analyses to explore the exposure-response relationship for brensocatib. The first analysis, presented in Report 00566-1 (2021), was based on data from the phase 2 study INS1007-201, which enrolled patients with non-cystic fibrosis bronchiectasis (NCFB). This model included 241 subjects, of whom 157 received brensocatib (either 10 mg or 25 mg once daily) and 84 received placebo. The PK component used a population model with two distributional compartments and linear clearance, with absorption described by a chain of six transit compartments and distinct absorption rates for fed and fasted states. For the PD component, the model evaluated two efficacy endpoints, pulmonary exacerbations and sputum neutrophil elastase (NE) activity, as well as adverse events of special interest (AESIs), including periodontal disease, hyperkeratosis, and non-pulmonary infections.

No significant relationship was observed between Brensocatib exposure (AUC_{0-24}) and pulmonary exacerbations. Likewise, sputum NE activity showed no meaningful correlation when analysed as a continuous variable. The applicant therefore used a categorical variable based on NE levels being below the limit of quantification (BLQ), which was not considered an adequate surrogate. Due to the limitations of the biomarker and the lack of consistent exposure-response relationships, this model provided limited support for dose selection.

In contrast, the second analysis (Report ICPD 00694-1, 2024) was based exclusively on phase 3 data from study INS1007-301, despite earlier plans to pool data from both phase 2 and 3. The rationale for excluding phase 2 data was not clearly justified, particularly given the 24-week duration of study 201. In the phase 3 model, sputum NE activity was excluded due to its weak performance in the prior model. Instead, the efficacy endpoints included annualized rate of pulmonary exacerbations, time to first exacerbation, and change from baseline in post-bronchodilator FEV1. Safety endpoints were limited to selected AESIs: hyperkeratosis, periodontal disease, and pneumonia.

This analysis included 1,189 patients from the intent-to-treat (ITT) population, of whom 626 received brensocatib (303 on 10 mg, 323 on 25 mg). The primary PK exposure variables were steady-state AUC24, Cmax, and Cmin. Statistically significant improvements in efficacy outcomes were observed with increasing AUC24. For both annualized pulmonary exacerbation rate and time to first exacerbation, an AUC24 threshold of 1100 ng·h/mL was found to be discriminatory; 30.7% of patients on 10 mg had exposures below this threshold, compared to none in the 25 mg group. For FEV1 response at Week 52, a higher threshold of 1531 ng·h/mL was identified, with 55.5% of patients on 10 mg falling below this

level, compared to only 0.6% in the 25 mg group.

The analyses suggested improved efficacy with higher exposure, and no corresponding increase in safety risks was identified. However, a slightly higher incidence of hyperkeratosis was noted in patients with the highest exposures, though all cases were mild or moderate. Overall, the submitted E-R analyses demonstrated a general trend of improved efficacy with greater exposure, but due to the reliance on a single phase 3 dataset and separate analyses of safety and efficacy, the regulatory impact of these models is considered limited.

Dose selection and therapeutic window

The applicant proposed a once-daily 25 mg dose as the clinically relevant regimen. During development, doses from 5 mg to 65 mg were evaluated, with dose selection informed by PK/PD modelling - an approach was deemed acceptable.

No significant covariate effects on brensocatib exposure were identified in clinical studies or the popPK model that would warrant dose adjustment.

In adolescents, exposure was slightly higher than in adults, potentially increasing the risk of adverse effects, though the overall risk appears similar between age groups.

6.2.6.2. Conclusions

Overall, it is concluded that the PK/PD profile of brensocatib has been reasonably characterized within the clinical programme. The metabolic profile of brensocatib has been adequately characterised in mass-balance study 103. The pharmacology and pharmacodynamic properties of brensocatib have been sufficiently investigated.

CHMP agrees that available clinical data in special populations (renal / hepatic impairment) do not warrant particular dose adaptations to be included in SmPC section 4.2. Equally, it is agreed that no relevant food effect was observed, i.e. brensocatib can be taken irrespective of meals.

Upon CHMP request, to further characterise the CYP 3A4 induction with brensocatib, the applicant committed to perform a post authorisation *in vivo* CYP3A4 induction study: to evaluate the effect of steady-state brensocatib on the pharmacokinetics of a sensitive CYP3A4 substrate, midazolam and 1-OH- midazolam in healty volunteers. The study will be completed in Q1 2028.

6.3. Clinical efficacy

6.3.1. Dose response studies

Prior to advancement to Phase 2, Phase 1 single dose and MAD studies in HP were conducted with single dose administration at 5 to 65 mg and repeated dosing at 10 to 40 mg QD. The clinical PK and PD data indicated that brensocatib provided adequate steady-state exposures at the proposed doses of 10 and 25 mg QD for further evaluation in the Phase 2 study in a population with NCFBE. The results of Phase 1 multiple ascending doses demonstrated further that plasma NE activity was reduced with increasing doses asymptotically approaching a flat relationship above 25 mg.

Based on the plasma NE activity and safety profile, 10 and 25 mg QD were selected to be tested in the Phase 2 Study INS1007-201 and in the Phase 3 Study INS1007-301.

6.3.2. Main study

6.3.2.1. INS1007-301

6.3.2.1.1. Study title

A Phase 3, Randomized, Double-Blind, Placebo-Controlled Study to Assess the Efficacy, Safety, and Tolerability of Brensocatib Administered Once Daily for 52 Weeks in Subjects with Non-Cystic Fibrosis Bronchiectasis - The ASPEN Study.

Table 60: Study identifiers and milestones

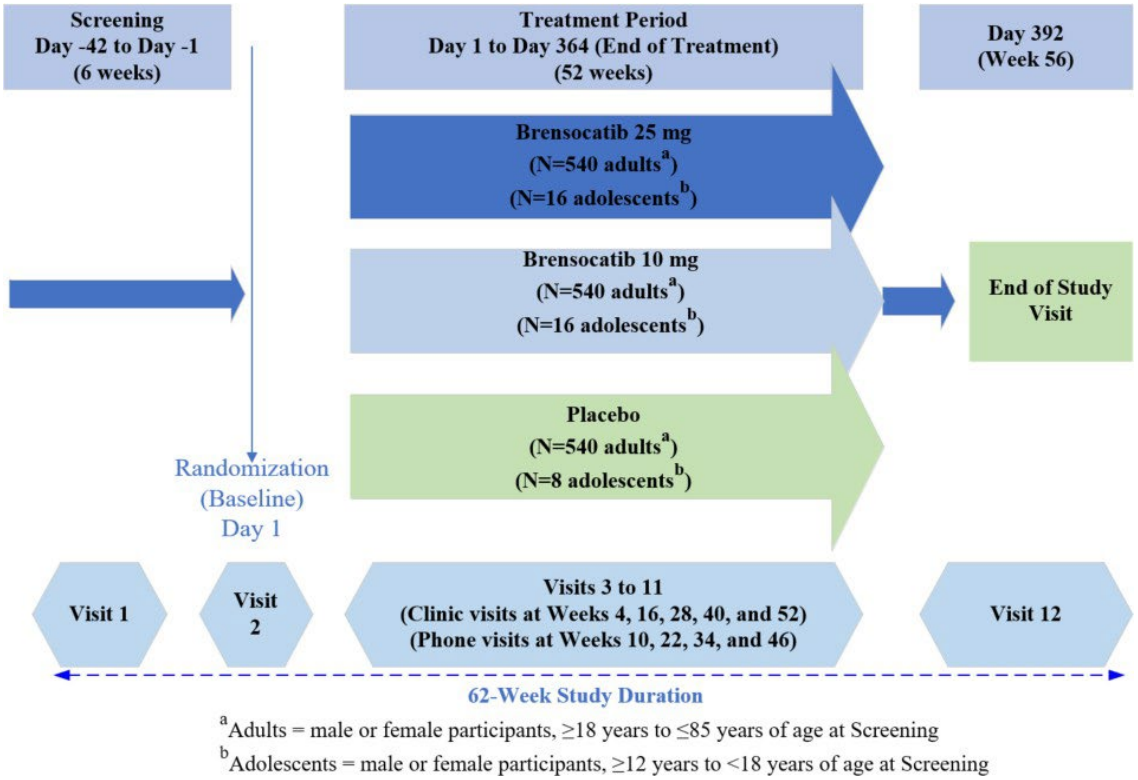
Study code	INS1007-301
EU CT number	2020-003688-25
NCT number	NCT04594369
ISRCT number	jRCT2031210048
Other identifier(s)	ASPEN Study
Location in eCTD	5.3.5.1
Sponsor:	Insmed Incorporated 700 US Highway 202/206 Bridgewater, NJ 08807-1704 USA
CSP Version 1.0	31 July 2020
Study Initiation Date:	10 November 2020 (first participant first visit)
Primary Completion Date:	28 March 2024 (data cutoff date, last adult participant completed the 52-week treatment period or discontinued from the study before Week 52)
SAP Final Version 1.0	19 April 2024
Primary database locked	15 May 2024
CSR Final Date:	20 September 2024

6.3.2.1.2. Study design

INS1007-301 (ASPEN) was a Phase 3, randomized, double-blind, placebo-controlled, parallel-group, multicentre, multinational study designed to evaluate the efficacy and safety of brensocatib 10 mg and 25 mg once daily in patients with non-cystic fibrosis bronchiectasis (NCFBE). The study had a 52-week treatment period followed by a 4-week follow-up period.

The Schedule of Assessments and Procedures is provided in the study protocol (Appendix 16.1.1, Global Amendment 4).

Figure 30: Study schema



Randomisation was stratified by baseline Pseudomonas aeruginosa colonisation status, number of prior pulmonary exacerbations (PEs), geographic region, and age group (adolescent vs adult).

An Independent Clinical Endpoint Committee (CEC) adjudicated all suspected PE events to determine whether they met the protocol-defined criteria.

Treatment

Participants received one tablet daily of either brensocatib 10 mg, 25 mg, or matching placebo for 52 weeks. Participants were instructed to take the medication with water at the same time each day, with or without food.

Table 61: Study Interventions Administered

Treatment:	**Placebo**	**Brensocatib**	
Description:	1 tablet QD × 52 weeks	1 tablet QD × 52 weeks	1 tablet QD × 52 weeks
Type:	Placebo	Drug	Drug
Formulation:	Tablet	Tablet	Tablet
Unit Dose:	NA	10 mg	25 mg
Route:	Oral	Oral	Oral
Use:	Experimental	Experimental	Experimental
Route:	Oral	Oral	Oral
IMP and NIMP:	IMP	IMP	IMP
Manufacturer:			
Sourcing:	Provided centrally by Sponsor		
Packaging and Labeling:	Provided in HDPE bottles and labeled as required per country requirements		

HDPE = high-density polyethylene, IMP = investigational medicinal product, NA = not applicable, NIMP = noninvestigational medicinal product, QD = once daily.

Randomisation

Randomisation was performed centrally using an interactive response technology system. Adult participants were randomized in a 1:1:1 ratio to receive brensocatib 10 mg, brensocatib 25 mg, or placebo. Randomizations were stratified based on the geographic region (North America, Europe, Japan, and the Rest of the World), sputum sample testing positive or negative for P. aeruginosa at Screening, and the number of PEs (2 or ≥3) in the previous 12 months.

Randomizations were enforced to have 30% of adults with ≥3 prior PEs, no more than 20% of participants >75 years of age, no more than 20% of adults with eosinophil count in peripheral blood ≥300/mm3 at Screening, and no more than 20% with COPD as a comorbidity.

Enrollment targets were established with up to 13% of adults from Eastern Europe and with North America, Western Europe, Asia Pacific, and Latin America contributing between 20% and 30% each. No single country (except the US) could contribute >15% of the overall randomized population.

Adolescent participants were randomized in a 2:2:1 ratio to the same treatment arms. There was no stratification for adolescents.

Blinding

The study employed a double-blind design. Participants, investigators, study staff, and the sponsor were blinded to treatment assignment throughout the study. The two brensocatib oral tablets 2 strengths 10- and 25-mg were round, biconvex, brown film-coated tablets, identical in size and appearance. Matching placebo tablets were identical to brensocatib film-coated tablets in shape, size, and colour.

Emergency unblinding was allowed only if knowledge of the treatment assignment was essential for the clinical management of the participant.

Patient population

Eligible participants were aged 12-85 years with HRCT-confirmed NCFBE and a history of ≥2 PEs requiring antibiotics in the prior 12 months (≥1 PE for adolescents).

A qualifying exacerbation was defined by the need for a physician-prescribed course of systemic antibiotics for signs and symptoms of respiratory infection, with no explicit requirement for a minimum number of symptoms or duration.

Key exclusion criteria included cystic fibrosis, active tuberculosis or non-tuberculous mycobacterial infection, and active smokers.

6.3.2.1.3. Objectives and estimands

Primary objective

To evaluate the effect of brensocatib on the annualized rate of adjudicated pulmonary exacerbations (PEs) over 52 weeks.

According to the CSP Amendment 4, a **pulmonary exacerbation** was defined by having ≥3 of the following symptoms for at least 48 hours, resulting in a physician's decision to prescribe systemic antibiotics:

- Increased cough
- Increased sputum volume or change in sputum consistency
- Increased sputum purulence
- Increased breathlessness and/or decreased exercise tolerance
- Fatigue and/or malaise
- Hemoptysis

Note: Subjects on chronic macrolide therapy whose only change in therapy is dose or frequency adjustment did not meet the definition of PE.

A **severe pulmonary exacerbation** was defined as those requiring IV antibacterial drug treatment and/or hospitalization.

Any pulmonary exacerbation in the study after randomization through the end of the 52-week treatment period was included in the primary endpoint calculation (provided it meets the protocol-defined criteria). A minimum of 14 days must occur between the end date of one pulmonary exacerbation and the start date of the next pulmonary exacerbation. Any exacerbations that occur less than 14 days from the prior exacerbation was not considered as a new exacerbation.

The time to the first pulmonary exacerbation was calculated from the randomization date to the onset date of the first exacerbation. Subjects who did not have an exacerbation at the end of the 52-week treatment period were censored at the date of Week 52.

The adjudication of pulmonary exacerbations was conducted by an independent, external CEC (see Section 3.3.1.1.5).

According to the CEC charter, a **pulmonary exacerbation** was defined by having ≥3 of the following symptoms for at least 48 hours, resulting in a physician's decision to prescribe systemic antibiotics:

• Increased cough

• Increased sputum volume or change in sputum consistency

• Increased sputum purulence

• Increased breathlessness and/or decreased exercise tolerance

• Fatigue and/or malaise

• Hemoptysis

A **severe pulmonary exacerbation** was defined as those requiring one the following at any point during a PE event:

• IV Antibiotics

• Hospitalization ≥ 24 hours

• Hospitalization ≥ 24 hours and IV Antibiotics

• Hospitalization < 24 hours and IV Antibiotics

A **Clinically Relevant Exacerbation** was defined as a non-severe PE event that requires prescription antibiotic treatment and did not meet the protocol definition of PE but was considered clinically relevant by the CEC.

The **PE Onset Date** was the date initial antibiotic treatment was prescribed, or the date initial antibiotic treatment was started when the prescribed date is not applicable and/or not available.

The **PE End Date** was the last day of antibiotic treatment associated with the resolution of a PE event, as determined by the CEC per clinical judgement and study protocol. The CEC performs a clinical assessment of the provided medical records and documentation related to each exacerbation. If the CEC determines that more than 1 course of antibiotic treatment applies to a single PE event, the PE end date will be the antibiotic stop date of the last course of antibiotics which correspond to the resolution of the event.

If the last day of antibiotic treatment was unclear or unable to be obtained, even after receiving a query response from the site, the PE end date was the date PE symptoms resolve. If both the last day of antibiotic treatment and the date PE symptoms resolve were unclear or unavailable, the PE End Date was considered *Unable to Determine*.

If the patient was hospitalized and outpatient antibiotic treatment was not prescribed at or after discharge, the PE end date was the last day of antibiotic administration; however, if this date was unavailable or the stop date/duration of antibiotic treatment was unclear, the PE end date was considered the date the patient was discharged from the hospital.

During CEC review, sequential courses of antibiotic treatment prescribed ≤ 14 days apart (i.e., between the stop date of 1 course and the prescribed date / start date of the following course), may be considered part of 1 individual PE event, per CEC assessment. Sequential courses of antibiotic treatment prescribed > 14 days apart were considered part of 2 separate PE events, per CEC assessment (see also "Changes in the planned conduct of the study").

Adjudication of PEs:

All 2,267 investigator-reported events were reviewed by the CEC. 1,897 (83.7%) were adjudicated as protocol-defined PEs, 212 (9.4%) as clinically relevant exacerbations (CREs), and 158 (7%) as not PEs. Only protocol-defined, CEC-adjudicated PEs were included in the primary and secondary efficacy

analyses ratio.

Statistical methods for estimation and sensitivity analysis on primary estimand

Estimands for the primary objective

Table 62: Primary Endpoint and Estimands

Variable	Population Level Summary	Estimand Name	Treatment Regimen[a]	ICEs/Strategy
Annualized rate of PEs while being exacerbation-free (as time while experiencing an exacerbation event was subtracted from the time at risk)	Rate ratio	Primary: On-Study	Assignment to IP regardless of discontinuation or modifications to standard of care	Early discontinuation from randomized IP/ Treatment Policy Modification to standard of care/ Treatment Policy
		Supplementary: On-Treatment	Standard of care + randomized IP	Early discontinuation from randomized IP/ While On-Treatment Modification to standard of care/ While On-Treatment
		Supplementary: On-IP	Randomized IP regardless of modifications to standard of care	Early discontinuation from randomized IP/ While On-Treatment Modification to standard of care/ Treatment Policy

a Randomized IP (ie, study treatment) includes brensocatib 10 mg QD, brensocatib 25 mg QD, or placebo QD in a 1:1:1 ratio for adult participants and a 2:2:1 ratio for adolescent participants.
Notes: Chronic treatment with antibiotics was permitted for all estimands if such treatment was initiated at least 3 months before Screening.
Modification to standard of care was defined as any change in chronic antibiotic use that was stable at baseline. This could have been any addition of a new chronic antibiotic or discontinuation from the participant's standard of care at baseline.
PE = pulmonary exacerbation, ICE = intercurrent event, IP = investigational product, QD = once daily.

The clinical question of interest is effect of brensocatib compared to placebo on annualised rate of PEs regardless of discontinuation of treatment or modifications to standard of care (expressed as rate ratio).

Secondary objectives

Key secondary objectives included:

- Time to first PE
- Proportion of participants who remained exacerbation-free during the treatment period
- Change in post-bronchodilator FEV1 (L) from baseline to Week 52

Postbronchodilator Pulmonary function tests (PFTs) by spirometry were performed per the ATS/ERS criteria and per the following instructions:

- When an inhaled SABA was used, 4 puffs of albuterol/salbutamol, levalbuterol/levosalbutamol, or terbutaline were administered. A postbronchodilator PFT was performed 15 to 30 minutes after the administration of albuterol or levalbuterol.
- When an inhaled SAMA was used, 4 puffs of ipratropium will be administered. A postbronchodilator PFT was performed within 20 to 30 minutes after the administration of ipratropium.
- If a subject could not perform an inhalation, the SABA or SAMA could be nebulized. Pulmonary function tests were performed within 20 to 30 minutes after finalization of nebulization.

    • Annualized rate of severe PE

- A severe pulmonary exacerbation was defined as those requiring IV antibacterial drug treatment and/or hospitalization.

    • Change in QOL-B Respiratory Symptoms Domain Score

The QOL-B is a multi-domain questionnaire designed specifically for patients with bronchiectasis. Each domain score represents a distinct aspect of health-related quality of life (HRQoL), such as:

- Respiratory Symptoms
- Physical Functioning
- Role Functioning
- Emotional Functioning
- Vitality
- Health Perceptions
- Social Functioning
- Treatment Burden

The QOL-B Respiratory Symptoms domain (Version 3.1) is part of the QOL-B, designed to assess patient-reported symptom burden in individuals with non-cystic fibrosis bronchiectasis.

The domain includes items focused on respiratory symptoms such as Cough, Sputum production, Chest congestion, Breathlessness. Responses are captured on Likert-type scales (e.g., frequency or severity). Each item contributes equally to the domain score.

Scores range from 0 to 100, with higher scores indicating better respiratory health (i.e., fewer or less severe symptoms).

In this study, a modified QOL-B was administered electronically using an eDiary system, with page 1 (demographics) removed since demographic information was captured elsewhere in the eCRF. The instrument was completed every 14 days from Day 1 (baseline) to Week 56. Validated translations in the local language were used where available. Subjects completed the questionnaire either in clinic (if the visit coincided with a scheduled QOL-B entry) or at home using the eDiary. Site staff were instructed to review completed questionnaires and retrain participants as needed.

Exploratory endpoints included other patient-reported outcomes (e.g., Bronchiectasis Exacerbation and Symptom (BEST) Tool, Patient Global Impression of Severity (PGI-S) scale, Patient Global Impression of Change (PGI-C) scale, EQ-5D-5L) and biomarker changes (e.g. neutrophil serine protease activity).

Estimands for the secondary objectives

Table 63: Seconday Endpoints and Estimands

Variable	Population Level Summary	Estimand Name	Treatment Regimen[a]	ICEs/Strategy
Time to first PE	Hazard Ratio	Primary : On-Study	Assignment to IP regardless of discontinuation or modifications to standard of care	Early discontinuation from randomized IP/Treatment Policy Modification to standard of care/ Treatment Policy
		Supplementary: On-IP	Randomized IP regardless of modifications to standard of care	Early discontinuation from randomized IP/While On-Treatment Modification to standard of care/ Treatment Policy
Responder status for exacerbation- free	Odds ratio	Primary : On-Study	Assignment to IP regardless of discontinuation or modifications to standard of care	Early discontinuation from randomized IP/Treatment Policy Modification to standard of care/ Treatment Policy
		Supplementary : Composite	Standard of care + randomized IP	Early discontinuation from randomized IP for lack of efficacy, tolerability, or death due to NCFBE/Composite (impute nonresponder) Early discontinuation from randomized IP for other reasons/Treatment Policy Early discontinuation from standard of care/Treatment Policy Addition of chronic antibiotics/ Composite (impute nonresponder)
Change in post-bronchodilator FEV_1 at Week 52	Mean difference	Primary : On-Study	Assignment to IP regardless of discontinuation or modifications to standard of care	Early discontinuation from randomized IP/Treatment Policy Modification to standard of care/ Treatment Policy
		Supplementary: On-IP	Randomized IP regardless of modifications to standard of care	Early discontinuation from randomized IP/While On-Treatment Modification to standard of care/ Treatment Policy

Annualized rate of severe PEs	Rate ratio	Primary : On-Study	Assignment to IP regardless of discontinuation or modifications to standard of care	Early discontinuation from randomized IP/Treatment Policy Modification to standard of care/ Treatment Policy
		Supplementary: On-IP	Randomized IP regardless of modifications to standard of care	Early discontinuation from randomized IP/While On-Treatment Modification to standard of care/ Treatment Policy
Change from baseline in QOL-B Respiratory Symptoms Domain Score at Week 52[b]	Mean difference	Primary : On-Study	Assignment to IP regardless of discontinuation or modifications to standard of care	Early discontinuation from randomized IP/Treatment Policy Modification to standard of care/ Treatment Policy
		Supplementary: On-IP	Randomized IP regardless of modifications to standard of care	Early discontinuation from randomized IP/While On-Treatment Modification to standard of care/ Treatment Policy

a Randomized IP (ie, study treatment) includes brensocatib 10 mg QD, brensocatib 25 mg QD, or placebo QD in a 1:1:1 ratio for adult participants and a 2:2:1 ratio for adolescent participants.
b The population for Change in QOL-B Respiratory Symptoms Domain Score at Week 52 was adult participants with NCFBE.
c The While On-Treatment reporting period included the 4-week follow-up post-discontinuation of randomized study treatment. Notes: Chronic treatment with antibiotics was permitted for all estimands if such treatment was initiated at least 3 months before Screening.
Modification to standard of care was defined as any change in chronic antibiotic use that was stable at baseline. This could have been addition of a new chronic antibiotic or discontinuation from the participant's standard of care at baseline.
FEV1 = forced expiratory volume in 1 second, ICE = intercurrent event, IP = investigational product, NCFBE = non-cystic fibrosis bronchiectasis, PE = pulmonary exacerbation, QD = once daily, QOL-B = Quality of Life Questionnaire – Bronchiectasis.

Statistical methods for estimation and sensitivity analysis

The Intent-to-Treat (ITT) Analysis Set comprises all participants who were randomized, excluding participants from Ukraine and participants from site USA065. This set was analyzed using the treatment to which the participant was randomized, regardless of the treatment actually received.

Efficacy analyses were primarily based on the ITT Analysis Set unless otherwise noted.

Primary analysis

The primary efficacy analysis evaluated the annualized rate of adjudicated pulmonary exacerbations (PEs) using a negative binomial regression model. The model included fixed effects for treatment group, Pseudomonas aeruginosa colonization status at screening (positive/negative), number of PEs in the prior 12 months (<3 vs ≥3), geographic region (North America, Europe, Japan, Rest of World), and age group (adolescent vs adult). The log of time at risk (excluding PE duration) was included as an offset variable.

The calculation of the time at risk as well the inclusion of data into the analysis depends on the underlying Estimand. Refer to Table below for further details. In general, participants' time at risk was the time on study excluding the time during exacerbations.

Table 64: PEs to be included into the analysis and time at risk based on Estimand

Estimand Name	PEs included in the analysis	Time at risk
On-Study (Primary)	All observed PEs up to Week 52 who meet the protocol defined criteria of PE diagnosis	Date of Week 52 visit / early study discontinuation[2] – date of randomization + 1 minus the sum of number of days the participant experiences exacerbation events that meet the protocol definition of PE diagnosis (end date of the PE – start date of the PE + 1). Refer to Section 6.7.1.1 for further instructions in case of combined PEs.
On-Treatment and On-IP (Supplementary)	All observed PEs up to end of IP / first ICE onset who meet the protocol defined criteria of PE diagnosis	Date of last IP intake / date of first ICE onset – date of first IP intake + 1 minus the sum of number of days the participant experiences exacerbation events that meet the protocol definition of PE diagnosis prior to the first ICE (end date of the PE – start date of the PE + 1). Refer to Section 6.7.1.1 for further instructions in case of combined PEs.

Secondary efficacy analyses

Key secondary endpoints were tested in a hierarchical sequence (gatekeeping procedure) to control the family-wise type I error rate at 5%. These endpoints included:

- Time to first PE (Cox proportional hazards model with the same covariates)
- Proportion of exacerbation-free participants (logistic regression)
- Change from baseline in post-bronchodilator FEV1 (repeated measures ANCOVA)
- Annualized rate of severe PEs (negative binomial regression)
- Change from baseline in QOL-B Respiratory Symptoms Domain Score (repeated measures ANCOVA)

All efficacy analyses were conducted in the intent-to-treat (ITT) population, which included all randomized participants excluding those from the Ukraine and one non-compliant site in the U.S.

Supportive and sensitivity analyses

Supportive analyses were conducted using alternative definitions of time at risk and statistical models. Sensitivity analyses included tipping-point and jump-to-reference methods to assess the robustness of missing data assumptions (MAR vs MNAR).

If there were study participants who discontinued randomized IP early and declined to remain in the study, ascertainment of data is incomplete. A "missing at random" (MAR) assumption for the missing data was made implicitly for the On-Study Estimand.

Robustness of the main analysis to departures from the MAR assumption were assessed using tipping point analyses. Due to the inherent difficulties in identifying the missing data mechanism in practice,

all missing data are assumed MNAR, and the tipping point penalties were applied accordingly. Tipping point analyses were also conducted for secondary endpoints.

A jump-to-reference sensitivity analysis was performed where the placebo arm acted as the reference arm. All data with an assessment or visit date prior to the time of the first of ICE was included. Data with an assessment or visit date after occurrence of an ICE related to treatment failure (eg, early discontinuation from randomized treatment for lack of efficacy, treatment discontinuation due to AE, or start of additional chronic antibiotics) was considered missing and imputed. Multiple imputations were used to replace missing outcomes for brensocatib- and placebo-treated study participants.

Exploratory analyses

Prespecified exploratory endpoints included other patient-reported outcomes (e.g. PGI-S, PGI-C, EQ-5D-5L) and pharmacodynamic biomarkers (e.g. NE activity). Subgroup analyses were conducted across demographic and clinical covariates, including age, sex, race, baseline eosinophil count, and maintenance macrolide use.

Planned subgroup analyses

Pre-specified subgroup analyses were planned to assess the consistency of treatment effects on key efficacy endpoints, particularly the primary endpoint (annualized rate of pulmonary exacerbations), across clinically relevant baseline characteristics.

The planned subgroups included:

- ☐ Age group: adolescent (12 to <18 years) vs adult (≥18 years)
- ☐ Sex: male vs female
- ☐ Race: White, Asian, Black or African American, and Other
- ☐ Geographic region: North America, Europe, Japan, and Rest of the World
- ☐ Baseline sputum Pseudomonas aeruginosa colonization: positive vs negative
- ☐ Number of PEs in the 12 months prior to screening: <3 vs ≥3
- ☐ Baseline post-bronchodilator FEV1 percent predicted: <50% vs ≥50%
- ☐ Bronchiectasis Severity Index (BSI) score categories: ≤4, 5–8, ≥9
- ☐ Maintenance use of macrolide antibiotics at baseline: yes vs no
- ☐ Use of inhaled corticosteroids at baseline: yes vs no
- ☐ Smoking history: never vs former smoker
- ☐ Baseline eosinophil count: <150, 150–<300, and ≥300 cells/μL
- ☐ History of asthma or COPD: yes vs no

For each subgroup, the treatment effect (rate ratio for PE rate or hazard ratio for time to first PE) was estimated along with corresponding 95% confidence intervals. Interaction terms between treatment and subgroup variables were included in exploratory models to assess heterogeneity of treatment effect.

The subgroup analyses were considered descriptive and not adjusted for multiplicity. Their primary purpose was to support consistency of efficacy across diverse patient populations and to inform benefit-risk considerations.

For adolescent subgroup (Age 12-<18), a Bayesian approach that borrows information from the adult subgroup (Age >= 18) was explored to inform efficacy on the primary endpoint. The prior distribution of the Bayesian approach was a mixture of a non-informative prior and a distribution of the treatment effects in adults, with the mixing weight ranging from 0 (no borrowing) to 1 (full borrowing) in increments of 0.05.

6.3.2.1.4. Results

Participant flow and numbers analysed

A total of 1767 participants were randomized in Study INS1007-301 across 36 countries and 460 study sites. Participants were allocated to one of three treatment groups:

- Brensocatib 10 mg: n = 589
- Brensocatib 25 mg: n = 587
- Placebo: n = 591

Following protocol-specified exclusions, the intent-to-treat (ITT) population consisted of 1721 participants. This excluded:

- 44 participants from Ukraine, due to early discontinuation following outbreak of war
- 2 participants from a U.S. site found to be non-compliant with GCP standards

Thus, the ITT set included:

- Brensocatib 10 mg: n = 583
- Brensocatib 25 mg: n = 575
- Placebo: n = 563

The Safety Analysis Set comprised all randomized participants who received at least one dose of study medication. Participants were analysed according to the treatment received.

- Brensocatib 10 mg: n = 663
- Brensocatib 25 mg: n = 663
- Placebo: n = 648

The Per Protocol (PP) population included participants from the ITT set who completed the study without major protocol deviations.

Participant disposition, including numbers randomized, treated, completed, and discontinued, along with reasons for discontinuation (e.g. adverse events, withdrawal of consent, loss to follow-up), were summarized and presented in the CSR. Most participants completed the 52-week treatment period, with balanced discontinuation rates across treatment arms.

Changes in the planned conduct of the study

Table 65: Protocol and amendments

Original Protocol	Version 1.0 - 31 July 2020	
Amendment 1	Version 4.0 - 12 March 2021	– To address health authority responses – To provide clarifications of study procedures
Amendment 2	Version 5.0 - 07 December 2021	– To add the adolescent population – (≥12 to <18 years of age) – To align with responses to health authority review comments
Amendment 3	Version 6.0 - 09 August 2022	– To add collection of PK blood and PD sputum samples from all study subjects, – To include the estimand framework – To clarify subjects from Ukraine will be replaced due to the war and that their data will be listed only and not included in the formal efficacy and safety analyses
Amendment 4	Version 7.0 - 13 February 2024	– To update the duration of the interval for defining a separate pulmonary exacerbation event, at least two weeks (14 days) must occur between the end date of an earlier PE and the start date of the next PE for the PEs to be considered separate events – To update the multiplicity control procedure of truncated Hochberg procedure to the enhanced mixture-based gatekeeping procedure with the primary endpoint and all secondary endpoints tested at alpha of 0.05 with primary endpoint also being tested at alpha of 0.01 – To add age group (adult, adolescent) as a potential covariate in the primary analysis model for the primary endpoint

Protocol amendment 4 (event definition):

A late protocol amendment (Version 7.0, 13 February 2024) introduced a 14-day rule for distinguishing separate PE events (≥14 days between end of one PE and start of the next, instead of ≥28 days). This rule was applied retrospectively to all 2,267 investigator-reported PEs, including previously adjudicated cases. Re-application of the new rule led to 57 events being split into 64, increasing the total to 2,331 PEs, of which 53 were protocol-defined. The CEC re-reviewed affected cases.

Baseline data

Baseline demographic and disease characteristics were well balanced across the three treatment groups in the intent-to-treat (ITT) population of Study INS1007-301 (N = 1721).

Age: The mean age was 60.2 years (range: 12 to 85 years), with approximately 48.8% of participants aged ≥65 years. A total of 41 adolescents (aged 12 to <18 years) were included in the study.

Sex: Females comprised 64.3% of the study population across treatment arms.

Race and ethnicity: The majority of participants were White (73.6%), followed by Asian (11.1%) and other racial groups. Most participants (67.5%) were not Hispanic or Latino.

Smoking status: Approximately 29.6% of participants were former smokers. The median pack-year history was approximately 9.9 across treatment arms.

Lung function: The mean post-bronchodilator percent predicted FEV1 (ppFEV1) at baseline was 73.5%, with 17.5% of participants having ppFEV1 <50%.

Disease severity and characteristics:

Mean Bronchiectasis Severity Index (BSI) score was 7.1, with 31.7% of participants classified in the severe category (BSI ≥9).

Approximately 35.3% of participants had baseline sputum cultures positive for Pseudomonas aeruginosa.

29.2% had experienced ≥3 PEs in the 12 months prior to screening.

24.5% had been hospitalized for a PE in the prior 24 months.

Concomitant treatments:

58.1% were using inhaled corticosteroids.

19.1% were on chronic macrolide therapy.

Outcomes and estimation - primary analysis (cut-off 28-Mar-2024)

Table 66: Summary of Multiplicity Controlled Endpoints (ITT Analysis Set)

Family	Brensocatib 10mg QD Treatment Effect	Brensocatib 25mg QD Treatment Effect	Brensocatib 10mg QD vs. Placebo p-value	Brensocatib 25mg QD vs. Placebo p-value	Brensocatib 10mg QD vs. Placebo Adjusted p-value*	Brensocatib 25mg QD vs. Placebo Adjusted p-value*
Family 1: Annualized rate of pulmonary exacerbations	0.789	0.806	0.0019	0.0046	0.0038	**0.0048**
Family 2: Time to first pulmonary exacerbation	0.813	0.825	0.0100	0.0182	0.0200	**0.0364**
Family 3: Responder status for exacerbation-free	1.412	1.400	0.0059	0.0074	0.0200	**0.0364**
Family 4: Change from baseline in post-bronchodilator FEV_1 at Week 52	0.011	0.038	0.3841	0.0054	0.3841	**0.0364**
Family 5: Annualized rate of severe pulmonary exacerbations	0.742	0.740	0.1277	0.1025	0.3841	**0.2050**
Family 6: Change from baseline in QOL-B Respiratory Symptoms Domain Score at Week 52	2.031	3.766	0.0594	0.0004	0.3841	**0.2050**

Source: CSR - Table 16.2.7.1-5
FEV1=Forced expiratory volume in 1 second, QOL-B=Quality of Life Questionnaire - Bronchiectasis.
Family 1 is of prime importance and consists of the two primary hypotheses H01: Brensocatib 10mg QD vs. Placebo and H02: Brensocatib 25mg QD vs. Placebo. All hypotheses related to secondary efficacy endpoints are grouped into Family 2 to Family 6.
*Adjusted p-values for multiplicity calculated using the enhanced mixture-based gatekeeping procedure. Family 1 was tested at two-sided alpha = 0.01 using Truncated Hochberg procedure with a truncation fraction of 0.9. Family 1 served as a gatekeeper for testing secondary endpoints sequentially in the order predefined in the statistical analysis plan.
Family 2 to Family 6 were tested at two-sided alpha=0.05.

Primary endpoint:

For the main analyses, the primary estimand was the On-Study Estimand in which all observed data up to Week 52 were included using the ITT Analysis Set.

Brensocatib significantly reduced the annualized rate of PEs compared with placebo by 21.1% in the 10 mg group and 19.4% in the 25 mg group. The annualized rate (95% CI) of PEs was 1.015 (0.910 to 1.132) in the 10 mg group, 1.036 (0.927 to 1.157) in the 25 mg group, and 1.286 (1.158 to 1.428) in the placebo group. The rate ratio (95% CI) for brensocatib compared with placebo was 0.789 (0.680 to 0.916) in the 10 mg group (P = 0.0019; adjusted P = 0.0038), and 0.806 (0.694 to 0.936) in the 25 mg group (P = 0.0046; adjusted P = 0.0048) (see table below).

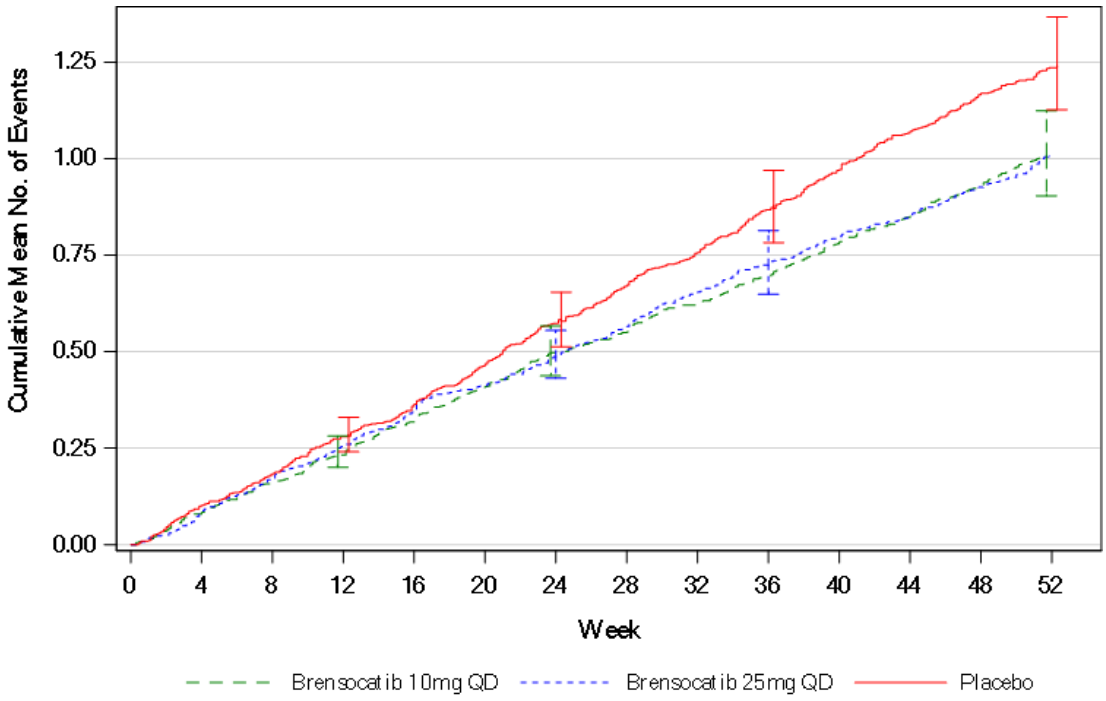
Table 67: Summary and Statistical Analysis of Primary Endpoint: Annualized Rate of Pulmonary Exacerbations - On-Study Estimand (ITT Analysis Set)

Characteristics	Brensocatib 10mg QD (N=583)	Brensocatib 25mg QD (N=575)	Placebo (N=563)
Participants with ≥1 exacerbation event, n (%)	292 (50.1)	288 (50.1)	324 (57.5)
Number of participants with exacerbation events, n (%)			
0	291 (49.9)	287 (49.9)	239 (42.5)
1	153 (26.2)	146 (25.4)	145 (25.8)
2	63 (10.8)	67 (11.7)	88 (15.6)
≥3	76 (13.0)	75 (13.0)	91 (16.2)
Total number of exacerbation events	563	554	669
Total time at risk in patient-years [1]	534.30	526.22	509.02
Annualized rate (95% CI) [2]	1.015 (0.910,1.132)	1.036 (0.927,1.157)	1.286 (1.158,1.428)
Rate ratio vs Placebo (95% CI) [2]	0.789 (0.680,0.916)	0.806 (0.694,0.936)	
p-Value [2]	0.0019	0.0046	
Total duration of exacerbation events (days) [3]	7445	7362	9094
n	563	554	669
Mean (SD)	13.2 (9.78)	13.3 (13.18)	13.6 (10.28)
Median	10.0	10.0	10.0
Min	1	1	1
Max	75	202	113

Source: CSR - Table 16.2.7.1-1
CI=Confidence interval, Min=Minimum, Max=Maximum, SD=Standard deviation, PE=Pulmonary exacerbation. A minimum of 2 weeks (14 days) must occur between the end date of an earlier PE and the start date of the next PE, otherwise, both PEs will be considered the same exacerbation.
Time at risk is calculated as: date of Week 52 or date of early study discontinuation - date of randomization + 1 minus the sum of number of days the participant experiences PE events that meet the protocol definition of PE diagnosis (end date of the PE - start date of the PE + 1).
Percentages are calculated based on the total number of participants in the analysis set per treatment as the denominator.
[1] The total time at risk of person-years is calculated by pooling all participants time at risk in years, separately for each treatment.
[2] Analysis of total number of exacerbation events is based on a negative binomial model including treatment group, sputum sample being classified as positive or negative for Pseudomonas aeruginosa at screening visit, the number of prior pulmonary exacerbations [<3 or ≥3] in the previous 12 months, stratification region (North America, Europe, Japan, and the Rest of World), and age group (adult, adolescent). The robust (empirical) sandwich estimator is used for the standard errors of the rates and rate ratios.
[3] PE events with no end date at Week 52 visit, the date of Week 52 visit is used as end date for the particular PE. The time at risk (natural log years) is the offset.

Over the course of the 52-week treatment period, participants in both brensocatib treatment groups had lower cumulative numbers of events compared with placebo (Figure below).

Figure 31: Cumulative Mean Number of Pulmonary Exacerbations Through Week 52 (ITT Analysis Set)



Number of Participants

Brensocatib 10mg QD	583	582	582	576	570	565	564	555	546	540	533	529	522	516
Brensocatib 25mg QD	575	572	568	566	563	552	550	543	540	537	528	523	520	515
Placebo	563	562	556	551	547	544	539	534	529	522	519	509	507	499

Source: C S R - Figure 16.2.7.1-2

ITT = intent-to-treat, No. = number, QD = once daily.

Key secondary endpoints

- **Time to First Pulmonary Exacerbation**

Brensocatib 10 and 25 mg significantly prolonged the time to first PE compared with placebo. The median (95% CI) time to first PE was 49.0 (40.0 to not estimable) weeks in the 10 mg group, 50.714 (37.571 to not estimable) weeks in the 25 mg group, and 36.714 (31.143 to 41.429) in the placebo group. The hazard ratio (95% CI) for brensocatib compared with placebo was 0.813 (0.695 to 0.952) in the 10 mg group (p = 0.0100; adjusted p = 0.0200) and 0.825 (0.703 to 0.968) in the 25 mg group (p = 0.0182; adjusted p = 0.0364).

Table 68: Time to First Pulmonary Exacerbation (ITT Analysis Set)

Characteristics	Brensocatib 10mg QD (N=583)	Brensocatib 25mg QD (N=575)	Placebo (N=563)
Number (%) of participants with any exacerbations	292 (50.1)	288 (50.1)	324 (57.5)
Kaplan-Meier estimates			
Median (Weeks or Days)	49.000	50.714	36.714
95% confidence interval	(40.000, NE)	(37.571, NE)	(31.143, 41.429)
Treatment comparison			
Hazard ratio point estimate	0.813	0.825	-
Hazard ratio 95% confidence interval	(0.695,0.952)	(0.703,0.968)	-
p-value	0.0100	0.0182	-
Adjusted p-value*	0.0200	0.0364	-

Source: CSP Table 16.2.7.2 1a.
ITT = intent-to-treat, NE = not estimable, PE = pulmonary exacerbation, QD = once daily.
For Study INS1007-301 the percentages are calculated based on the total number of participants in the analysis set per treatment as the denominator. Time to first PE is calculated as date of first exacerbation - date of randomisation + 1.
The hazard ratio estimates are obtained from the Cox proportional hazard model including effects for treatment, sputum sample being classified as positive or negative for Pseudomonas aeruginosa at Screening Visit, the number of prior PEs [<3 or ≥3] in the previous 12 months, stratification region (North America, Europe, Japan, and the Rest of the World), and age group (adult, adolescent). The robust sandwich estimator is used to estimate the covariance matrix.
A hazard ratio <1.0 indicates a lower average risk and a longer time to PE for brensocatib relative to placebo.
*Adjusted p-values for multiplicity calculated using the enhanced mixture-based gatekeeping procedure. Family 1 was tested at two-sided alpha = 0.01 using Truncated Hochberg procedure with a truncation fraction of 0.9. Family 1 served as a gatekeeper for testing secondary endpoints sequentially in the order predefined in the statistical analysis plan. Family 2 to Family 6 were tested at two-sided alpha=0.05.

- **Responder status for exacerbation-free over 52 weeks**

Participants in each brensocatib group were more likely to achieve exacerbation-free status over the 52-week treatment period compared with placebo. The brensocatib 10 and 25 mg groups had a higher proportion of responders compared with placebo (48.5% and 48.5% versus 40.3%, respectively).Treatment group differences compared with placebo were statistically significant for the brensocatib 10 mg dose (OR [95% CI]: 1.412 [1.105 to 1.806]; p = 0.0059 and adjusted p = 0.0200) and the brensocatib 25 mg dose (OR [95% CI]: 1.400 [1.095 to 1.792]; p = 0.0074 and adjusted p = 0.0364). Results from supplementary analysis using the Composite estimand were consistent with the primary analysis.

Table 69: Status of Exacerbation-Free Over the 52-Week Treatment Period – On-Study Estimand (ITT Analysis Set) (ITT Analysis Set)

	Brensocatib 10mg QD (N=583)	Brensocatib 25mg QD (N=575)	Placebo (N=563)
Responders, Average of 100 imputed datasets, n (%)	282.9 (48.5)	278.7 (48.5)	226.7 (40.3)
Non-Responders, Average of 100 imputed datasets, n (%)	300.1 (51.5)	296.3 (51.5)	336.3 (59.7)
At least one confirmed PE (pre-imputation)	292 (50.1)	288 (50.1)	324 (57.5)
Treatment comparison: Brensocatib vs Placebo			
Odds ratio	1.412	1.400	-
Odds ratio 95% Wald confidence interval	(1.105,1.806)	(1.095,1.792)	-
p-value	0.0059	0.0074	-
Adjusted p-value*	0.0200	0.0364	-

ITT=intent-to-treat, PE=pulmonary exacerbation, QD=once daily.
Percentages are calculated based on the total number of participants in the analysis set per treatment as the denominator.
Participants are considered responders if they complete 52-weeks on-study and have no protocol-defined PEs based on independent adjudication process up to the 52 Week follow-up period, or are censored earlier than Week 52 and imputed with ≥365 days.
Analysis based on 100 logistic regression models, including treatment group, sputum sample being classified as positive or negative for Pseudomonas aeruginosa at Screening Visit, the number of PEs [<3 or ≥3] in the previous 12 months, stratification region (North America, Europe, Japan, and the Rest of the World), and age group (adult, adolescent) as effects. The parameter estimates are combined using Rubin's rules and then exponentiated to show the odds ratio.
*Adjusted p-values for multiplicity calculated using the enhanced mixture-based gatekeeping procedure. Family 1 was tested at two-sided alpha = 0.01 using Truncated Hochberg procedure with a truncation fraction of 0.9. Family 1 served as a gatekeeper for testing secondary endpoints sequentially in the order predefined in the statistical analysis plan. Family 2 to Family 6 were tested at two-sided alpha=0.05.

- **Change in Post-bronchodilator FEV1**

Brensocatib demonstrated a consistent reduction of loss lung function as measured by the change from baseline in post-bronchodilator FEV_1. The effect reached statistical significance with brensocatib 25 mg (p = 0.0054 vs placebo). Brensocatib 25 mg showed a statistically significant lower reduction in post-bronchodilator FEV_1 at Week 52 in comparison to placebo with an LS mean (SE) difference of 38 (13.6) mL. Brensocatib 10 mg showed a lower reduction vs placebo with an LS mean (SE) difference of 11 (13.2) mL.

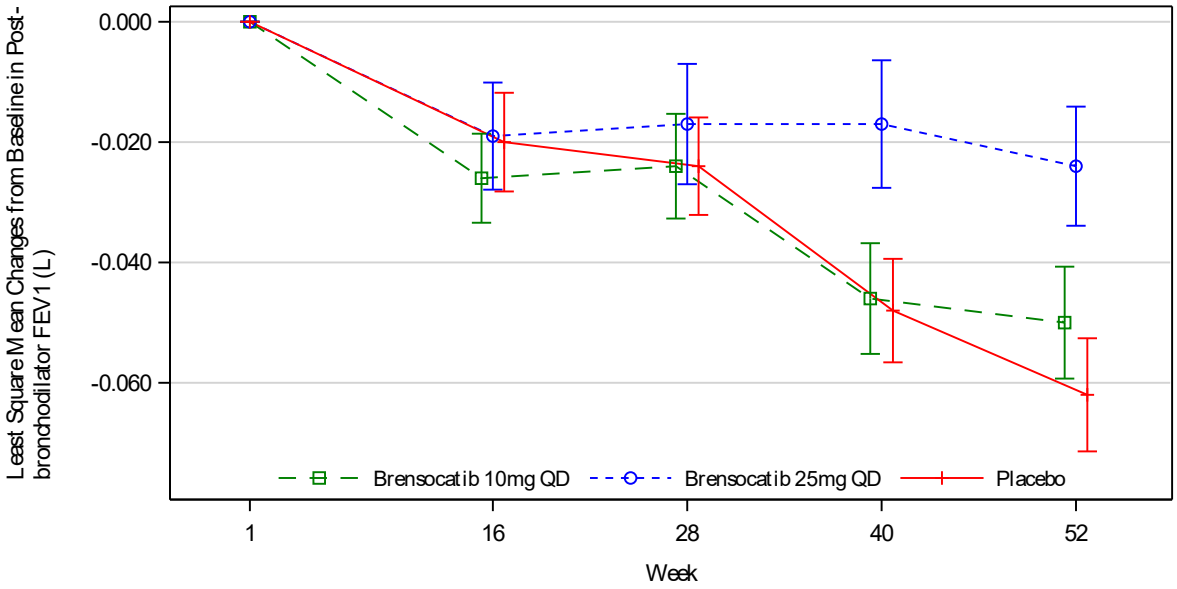
Table 70: Change in FEV1 (ITT Analysis Set)

Endpoint	Brensocatib 10 mg (N=583)	Brensocatib 25 mg (N=575)	Placebo (N=563)
Number of participants in model	564	551	539
LS mean (SE) (L)	-0.050 (0.0093)	-0.024 (0.0099)	-0.062 (0.0094)
LS mean difference (SE) (L)	0.011 (0.0132)	0.038 (0.0136)	--
p-value	0.3841	0.0054	--
Adjusted p-value*	0.3841	0.0364	--

CI = confidence interval, FEV_1 = forced expiratory volume in 1 second, L = litres, LS = Least Squares, PE = pulmonary exacerbation, SE = standard error.
*Adjusted p-values for multiplicity calculated using the enhanced mixture-based gatekeeping procedure. Family 1 was tested at two-sided alpha = 0.01 using Truncated Hochberg procedure with a truncation fraction of 0.9. Family 1 served as a gatekeeper for testing secondary endpoints sequentially in the order predefined in the statistical analysis plan. Family 2 to Family 6 were tested at two-sided alpha=0.05.

Patients with NCFBE experience a decline in lung function that has been estimated to be over 50 mL per year in FEV_1 versus around 30 mL per year in healthy non-smoking adults after age 30 (Thomas et al., 2019). In Study INS1007-301, placebo and brensocatib 10 mg showed a decrease in FEV_1 of 62 mL and 50 mL respectively. In comparison, brensocatib 25 mg showed a 24 mL decrease in FEV_1, suggesting a therapeutic effect in terms of lung function decline. Moreover, as shown in the figure below, FEV_1 remained stable with brensocatib 25 mg after Week 16, in contrast to brensocatib 10 mg and placebo groups, that showed lung function decline consistent with that described in the NCFBE population.

Figure 32: Least Squares Mean Changes (± SE) from Baseline in Post-Bronchodilator FEV1 (L) Over Time – (ITT Analysis Set)



Number of Participants with Observation					
Brensocatib 10mg QD	579	545	529	513	475
Brensocatib 25mg QD	571	529	523	494	487
Placebo	563	522	513	494	468

FEV_1 = forced expiratory volume in 1 second; QD = once daily.
Baseline is the most recent non-missing assessment determined as best effort prior to the first dose of the investigational product.
Only best efforts were used in the statistical analysis.

- **Annualised Rate of Severe Pulmonary Exacerbations**

In Study INS1007-301, a reduction in the annualised rate of severe PEs compared to placebo was observed for both doses. The magnitude of effect was similar between the 2 doses and comparable to that seen for the overall rate of PEs. The annualised rate (95% CI) of severe PEs was 0.137 (0.103 to 0.182) in the 10 mg group, 0.137 (0.105 to 0.179) in the 25 mg group, and 0.185 (0.142 to 0.242) in the placebo group. The rate ratio (95% CI) for brensocatib compared with placebo was 0.742 (0.505 to 1.089) in the 10 mg group (nominal p = 0.1277) and 0.740 (0.515 to 1.062) in the 25 mg group (nominal p = 0.1025).

Table 71: Annualised Rate of Severe PEs (ITT Analysis Set)

Endpoint	Brensocatib 10 mg (N=583)	Brensocatib 25 mg (N=575)	Placebo (N=563)
Total number of exacerbation events	86	80	109
Annualised rate (95% CI)[a]	0.137 (0.103,0.182)	0.137 (0.105,0.179)	0.185 (0.142,0.242)
Rate ratio vs Placebo (95% CI)[a]	0.742 (0.505,1.089)	0.740 (0.515,1.062)	--
p-value	0.1277	0.1025	--
Adjusted p-value*	0.3841	0.2050	--

CI = confidence interval, ITT = intent-to-treat, PE = pulmonary exacerbation, QD = once daily.
For Study INS1007-301: Percentages are calculated based on the total number of participants in the analysis set per treatment as the denominator.
[a] For Study INS1007-301 the analysis of total number of exacerbation events is based on a negative binomial model including treatment group, sputum sample being classified as positive or negative for Pseudomonas aeruginosa at Screening Visit, the number of prior pulmonary exacerbations [<3 or ≥3] in the previous 12 months, stratification region (North America, Europe, Japan, and the Rest of World), and age group (adult, adolescent). The robust (empirical) sandwich estimator is used for the standard errors of the rates and rate ratios.
*Adjusted p-values for multiplicity calculated using the enhanced mixture-based gatekeeping procedure. Family 1 was tested at two-sided alpha = 0.01 using Truncated Hochberg procedure with a truncation fraction of 0.9. Family 1 served as a gatekeeper for testing secondary endpoints sequentially in the order predefined in the statistical analysis plan. Family 2 to Family 6 were tested at two-sided alpha=0.05.

- **Change in QOL-B Respiratory Symptoms Domain Score**

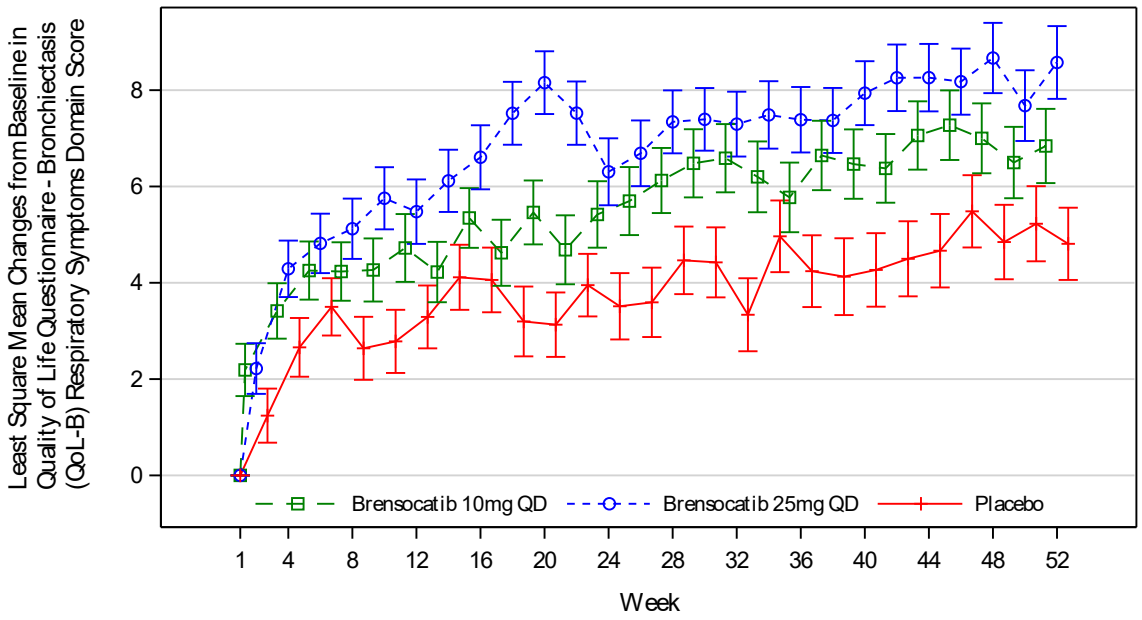
Brensocatib 25 mg demonstrated nominally significant improvements in QOL-B Respiratory Symptoms Domain Score versus placebo at Week 52, while brensocatib 10 mg showed numerical improvements compared to placebo. The LS mean (SE) change from Baseline at Week 52 was 6.841 (0.7706) in the 10 mg group, 8.575 (0.7556) in the 25 mg group, and 4.809 (0.7500) in the placebo group. The LS mean (SE) difference in the change from Baseline for brensocatib compared with placebo was 2.031 (1.0775) in the 10 mg group (nominal p = 0.0594) and 3.766 (1.0642) in the 25 mg group (nominal p = 0.0004).

Table 72: Change in QOL-B Respiratory Symptoms Domain Score at Week 52a (adults only) (ITT Analysis Set)

Endpoint	Brensocatib 10 mg (N=583)	Brensocatib 25 mg (N=575)	Placebo (N=563)
Number of participants in model	487	495	486
LS mean (SE)	6.841 (0.7706)	8.575 (0.7556)	4.809 (0.7500)
LS mean difference (SE)	2.031 (1.0775)	3.766 (1.0642)	--
p-value	0.0594	0.0004	--
Adjusted p-value*	0.3841	0.2050	--

CI = confidence interval, LS = Least Squares, QD = once daily, QOL-B = quality of life questionnaire-bronchiectasis, SD = standard deviation, SE = standard error.
[a] For Study INS1007-301 the analysis is based on a linear repeated measures model including treatment group, visit, treatment-by-visit interaction, sputum sample being classified as positive or negative for Pseudomonas aeruginosa at Screening Visit, the number of pulmonary exacerbations [<3 or ≥3] in the previous 12 months, and stratification region (North America, Europe, Japan, and the Rest of the World) as fixed effects, and baseline value as covariate. The variance-covariance structure is compound symmetric with the robust sandwich variance estimator.
*Adjusted p-values for multiplicity calculated using the enhanced mixture-based gatekeeping procedure. Family 1 was tested at two-sided alpha = 0.01 using Truncated Hochberg procedure with a truncation fraction of 0.9. Family 1 served as a gatekeeper for testing secondary endpoints sequentially in the order predefined in the statistical analysis plan. Family 2 to Family 6 were tested at two-sided alpha=0.05.

Figure 33: LS Mean Changes (SE from Baseline in QOL-B Respiratory Symptoms) Domain Score Over Time (ITT Analysis Set)



Number of Participants with Observation

Brensocatib 10mg QD	488	473	463	456	453	450	446	443	418	418	435	423	416	381
Brensocatib 25mg QD	497	476	464	459	454	456	442	446	434	438	432	426	423	394
Placebo	487	457	459	452	448	437	421	434	428	411	408	405	399	366

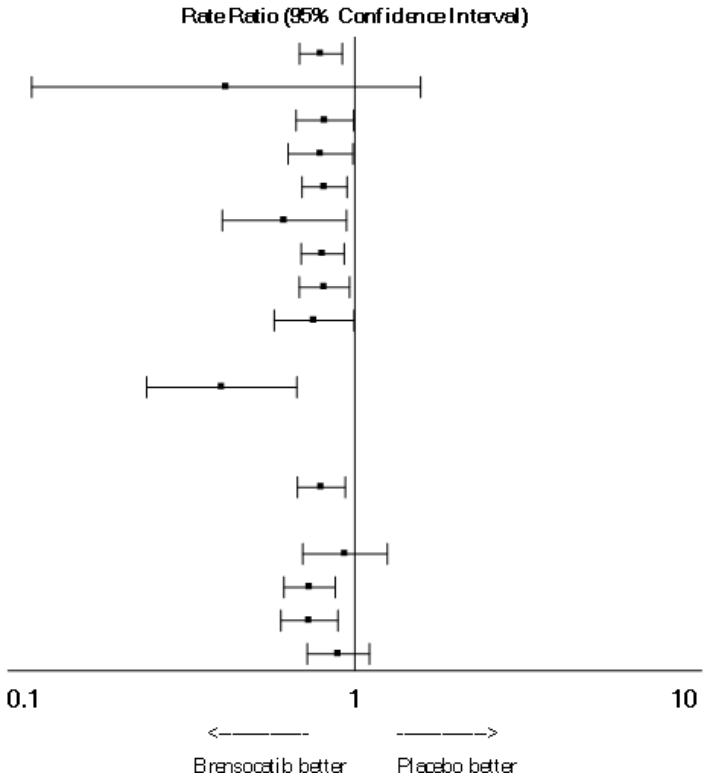
ITT = intent-to-treat, LS = Least Squares; QD = once daily, QOL-B = quality of life questionnaire-bronchiectasis, SE = standard error.
Baseline is the most recent non-missing value prior to the first dose of the investigational product. Week 1 is referring to the baseline assessment.

Pre-defined and post-hoc subgroup analyses

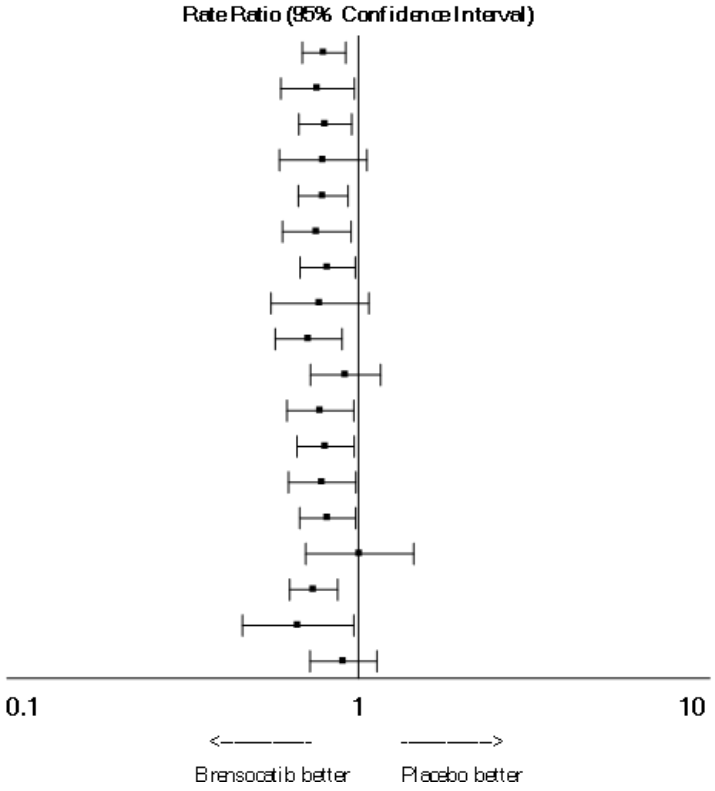
Pre-specified subgroup analyses were conducted for the primary endpoint (annualized PE rate) to assess the consistency of treatment effects across clinically relevant patient groups. These analyses were descriptive and not adjusted for multiplicity.

Across subgroups, the treatment effect of brensocatib (both 10 mg and 25 mg) on PE reduction was generally consistent. There was no evidence of differential efficacy based on age, sex, or geographic region. The benefit of brensocatib 25 mg was observed consistently across subgroups, including in those with high disease burden (e.g. BSI ≥9, P. aeruginosa colonisation, FEV1 <50%) (see figures below).

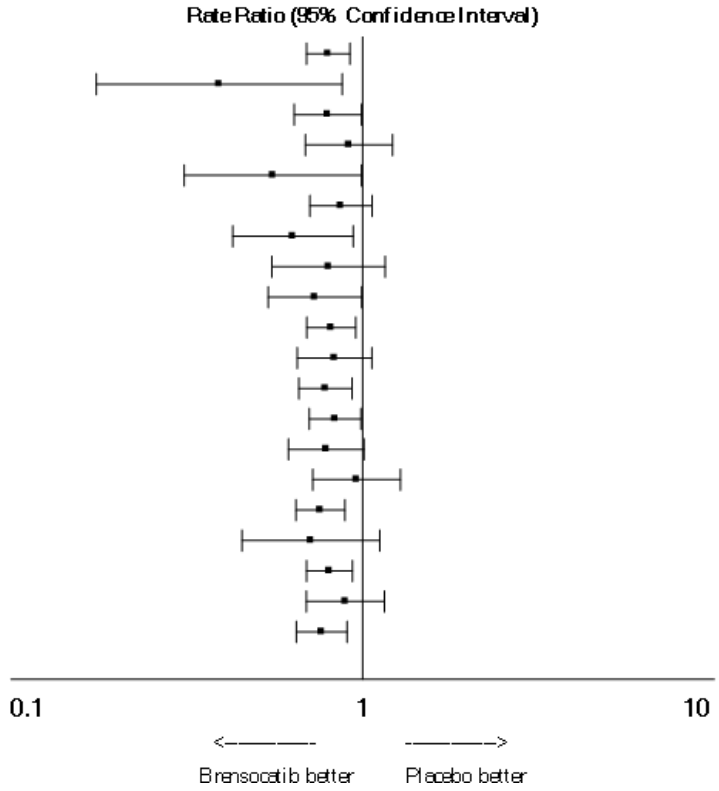
Figure 34: Plot of Primary Endpoint: Forest Plot of Annualized Rate of Pulmonary Exacerbations for Brensocatib 10 mg QD vs Placebo - On-Study Estimand (ITT Analysis Set)



Subgroup	10mg QD (N=583) n (%)	Placebo (N=563) n (%)	Rate Ratio (95% CI)
Overall	583 (100.0)	563 (100.0)	0.789 (0.680, 0.916)
Age: 12 to <18 years	17 (2.9)	8 (1.4)	0.408 (0.105, 1.582)
Age: 18 to <65 years	289 (49.6)	295 (52.4)	0.812 (0.664, 0.992)
Age: >=65 years	277 (47.5)	260 (46.2)	0.788 (0.628, 0.987)
Age: <75 years	500 (85.8)	470 (83.5)	0.810 (0.692, 0.948)
Age: >=75 years	83 (14.2)	93 (16.5)	0.612 (0.397, 0.944)
Age: >=18 years	566 (97.1)	555 (98.6)	0.799 (0.688, 0.929)
Sex: Female	385 (66.0)	362 (64.3)	0.809 (0.679, 0.964)
Sex: Male	198 (34.0)	201 (35.7)	0.753 (0.570, 0.995)
Race: American Indian or Alaska Native	8 (1.4)	9 (1.6)	NE
Race: Asian	63 (10.8)	64 (11.4)	0.396 (0.234, 0.668)
Race: Black or African American	2 (0.3)	3 (0.5)	NE
Race: Native Hawaiian or Other Pacific Islander	1 (0.2)	1 (0.2)	NE
Race: White	431 (73.9)	405 (71.9)	0.792 (0.670, 0.936)
Race: Other	15 (2.6)	11 (2.0)	NE
Ethnicity: Hispanic or Latino	177 (30.4)	170 (30.2)	0.935 (0.696, 1.255)
Ethnicity: Not Hispanic or Latino	391 (67.1)	373 (66.3)	0.730 (0.610, 0.873)
Number of PEs in prior 12 months: <3	411 (70.5)	396 (70.3)	0.728 (0.597, 0.889)
Number of PEs in prior 12 months: >=3	172 (29.5)	167 (29.7)	0.892 (0.718, 1.108)



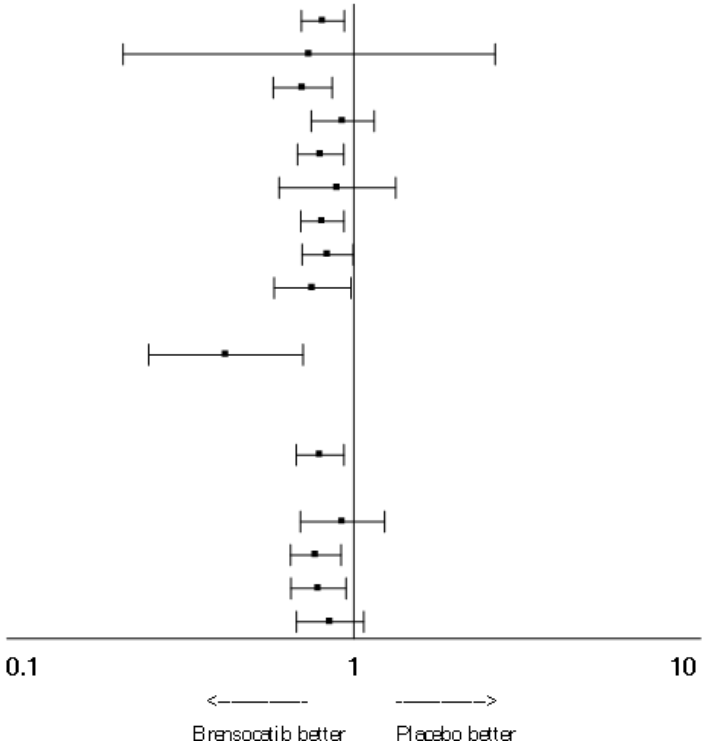
Subgroup	Brensocatib 10mg QD (N=583) n (%)	Placebo (N=563) n (%)	Rate Ratio (95% CI)
Overall	583 (100.0)	563 (100.0)	0.789 (0.680, 0.916)
Chronic use of antibiotics at baseline: Yes	146 (25.0)	133 (23.6)	0.755 (0.586, 0.973)
Chronic use of antibiotics at baseline: No	437 (75.0)	430 (76.4)	0.797 (0.664, 0.955)
Maintenance use of macrolides at baseline: Yes	110 (18.9)	105 (18.7)	0.785 (0.581, 1.061)
Maintenance use of macrolides at baseline: No	473 (81.1)	458 (81.3)	0.784 (0.681, 0.931)
P. aeruginosa colonization at baseline: Positive	203 (34.8)	199 (35.3)	0.751 (0.593, 0.951)
P. aeruginosa colonization at baseline: Negative	380 (65.2)	364 (64.7)	0.810 (0.689, 0.979)
Baseline BSI score: <=4	136 (23.3)	148 (26.3)	0.767 (0.548, 1.074)
Baseline BSI score: 5-8	275 (47.2)	220 (39.1)	0.710 (0.564, 0.894)
Baseline BSI score: >=9	168 (28.8)	195 (34.6)	0.916 (0.719, 1.166)
Baseline BSI score: <Median	286 (49.1)	275 (48.8)	0.770 (0.612, 0.969)
Baseline BSI score: >=Median	293 (50.3)	288 (51.2)	0.798 (0.655, 0.972)
BE-CT score: <Median	274 (47.0)	255 (45.3)	0.780 (0.618, 0.983)
BE-CT score: >=Median	309 (53.0)	308 (54.7)	0.810 (0.668, 0.982)
Baseline FEV1 % predicted post-bronchodilator: <50%	101 (17.3)	98 (17.4)	1.009 (0.696, 1.464)
Baseline FEV1 % predicted post-bronchodilator: >=50%	478 (82.0)	465 (82.6)	0.735 (0.623, 0.867)
Stratification region: North America	84 (14.4)	81 (14.4)	0.660 (0.450, 0.969)
Stratification region: Europe	231 (39.6)	221 (39.3)	0.903 (0.717, 1.136)



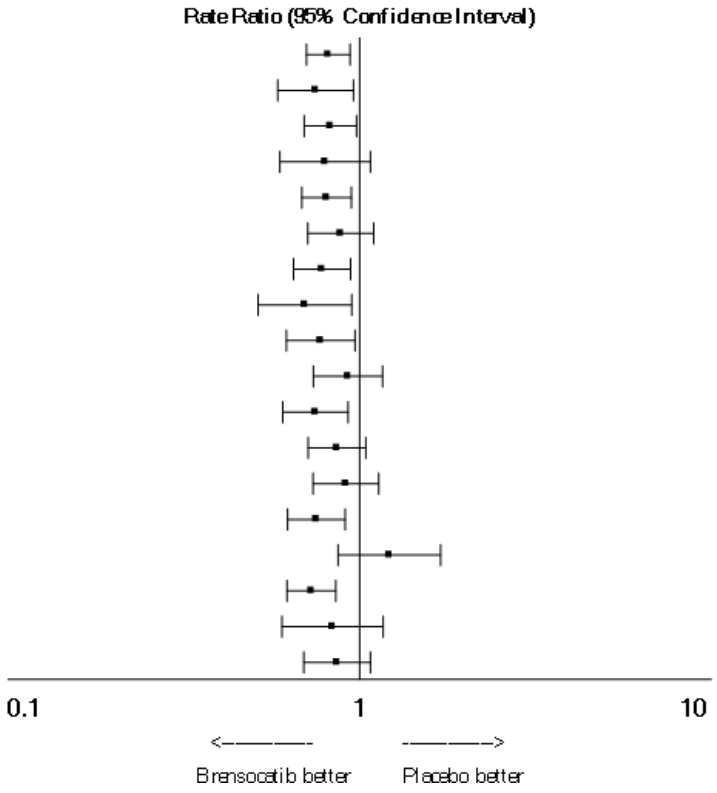
Subgroup	Brensocatib 10mg QD (N=583) n (%)	Placebo (N=563) n (%)	Rate Ratio (95% CI)
Overall	583 (100.0)	563 (100.0)	0.789 (0.680, 0.916)
Stratification region: Japan	30 (5.1)	29 (5.2)	0.373 (0.160, 0.869)
Stratification region: Rest of World	238 (40.8)	232 (41.2)	0.787 (0.625, 0.992)
Geographic region: South America	174 (29.8)	157 (27.9)	0.911 (0.675, 1.229)
Geographic region: Eastern Europe	75 (12.9)	72 (12.8)	0.540 (0.293, 0.992)
Geographic region: Western Countries	216 (37.0)	201 (35.7)	0.861 (0.696, 1.067)
Geographic region: Asian Countries	81 (13.9)	88 (15.6)	0.619 (0.409, 0.938)
Geographic region: Oceania	37 (6.3)	45 (8.0)	0.792 (0.536, 1.169)
Baseline eosinophil count: >=300/mm3	115 (19.7)	106 (18.8)	0.720 (0.522, 0.994)
Baseline eosinophil count: <300/mm3	465 (79.8)	452 (80.3)	0.806 (0.681, 0.954)
Smoking status: Former smoker	164 (28.1)	183 (32.5)	0.825 (0.638, 1.066)
Smoking status: Never smoked	419 (71.9)	380 (67.5)	0.775 (0.645, 0.930)
Use of inhaled steroids: Yes	324 (55.6)	352 (62.5)	0.828 (0.693, 0.989)
Use of inhaled steroids: No	259 (44.4)	211 (37.5)	0.779 (0.600, 1.010)
History of asthma: Yes	101 (17.3)	111 (19.7)	0.960 (0.710, 1.298)
History of asthma: No	482 (82.7)	452 (80.3)	0.748 (0.632, 0.886)
History of COPD: Yes	77 (13.2)	102 (18.1)	0.701 (0.436, 1.126)
History of COPD: No	506 (86.8)	461 (81.9)	0.796 (0.680, 0.933)
Hospitalized in prior 24 months for PE: Yes	146 (25.0)	142 (25.2)	0.889 (0.679, 1.162)
Hospitalized in prior 24 months for PE: No	437 (75.0)	421 (74.8)	0.755 (0.634, 0.899)

Note: Protocol-defined PEs are based on the independent adjudication process.
BE-CT = Bronchiectasis-Computed Tomography, BSI = Bronchiectasis Severity Index, CI = confidence interval, COPD = chronic obstructive pulmonary disease, CT = computed tomography, ITT = intent-to-treat, FEV1 = forced expiratory volume in 1 second, NE = not estimable, PE = pulmonary exacerbation, QD = once daily.

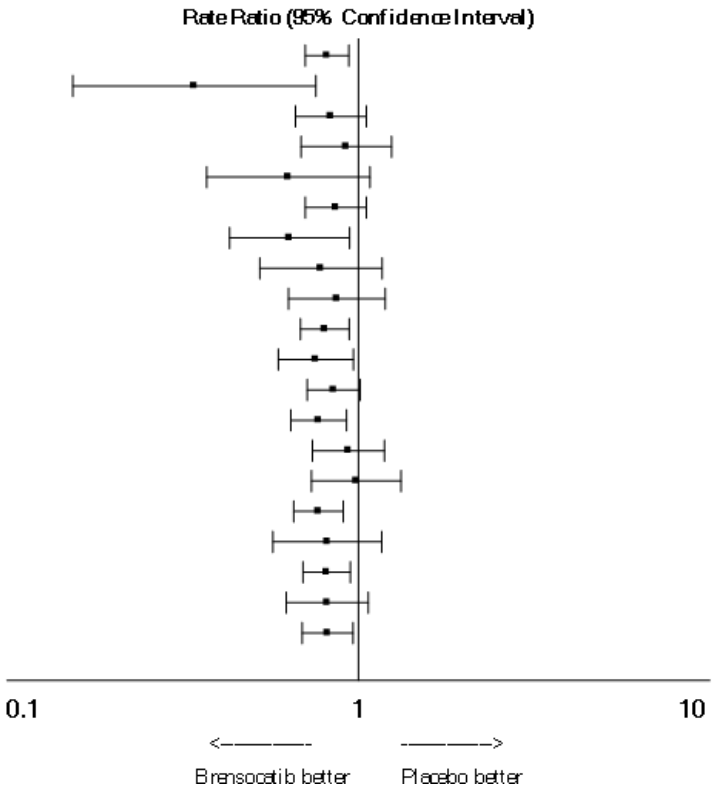
Figure 35: Plot of Primary Endpoint: Forest Plot of Annualized Rate of Pulmonary Exacerbations for Brensocatib 25 mg QD vs Placebo - On-Study Estimand (ITT Analysis Set)



Subgroup	(N=575) n (%)	(N=563) n (%)	Rate Ratio (95% CI)
Overall	575 (100.0)	563 (100.0)	0.806 (0.694, 0.936)
Age: 12 to <18 years	16 (2.8)	8 (1.4)	0.733 (0.200, 2.681)
Age: 18 to <65 years	257 (44.7)	295 (52.4)	0.700 (0.570, 0.860)
Age: >=65 years	302 (52.5)	260 (46.2)	0.926 (0.743, 1.153)
Age: <75 years	491 (85.4)	470 (83.5)	0.794 (0.676, 0.932)
Age: >=75 years	84 (14.6)	93 (16.5)	0.892 (0.594, 1.339)
Age: >=18 years	559 (97.2)	555 (98.6)	0.803 (0.691, 0.934)
Sex: Female	360 (62.6)	362 (64.3)	0.834 (0.697, 0.997)
Sex: Male	215 (37.4)	201 (35.7)	0.750 (0.574, 0.981)
Race: American Indian or Alaska Native	6 (1.0)	9 (1.6)	NE
Race: Asian	64 (11.1)	64 (11.4)	0.410 (0.239, 0.702)
Race: Black or African American	5 (0.9)	3 (0.5)	NE
Race: Native Hawaiian or Other Pacific Islander	0 (0.0)	1 (0.2)	NE
Race: White	430 (74.8)	405 (71.9)	0.790 (0.669, 0.934)
Race: Other	13 (2.3)	11 (2.0)	NE
Ethnicity: Hispanic or Latino	164 (28.5)	170 (30.2)	0.924 (0.689, 1.240)
Ethnicity: Not Hispanic or Latino	397 (69.0)	373 (66.3)	0.767 (0.643, 0.915)
Number of PEs in prior 12 months: <3	412 (71.7)	396 (70.3)	0.782 (0.645, 0.948)
Number of PEs in prior 12 months: >=3	163 (28.3)	167 (29.7)	0.848 (0.670, 1.072)



Subgroup	Brensocatib 25mg QD (N=575) n (%)	Placebo (N=563) n (%)	Rate Ratio (95% CI)
Overall	575 (100.0)	563 (100.0)	0.806 (0.694, 0.936)
Chronic use of antibiotics at baseline: Yes	154 (26.8)	133 (23.6)	0.739 (0.569, 0.959)
Chronic use of antibiotics at baseline: No	421 (73.2)	430 (76.4)	0.818 (0.683, 0.980)
Maintenance use of macrolides at baseline: Yes	114 (19.8)	105 (18.7)	0.789 (0.578, 1.077)
Maintenance use of macrolides at baseline: No	461 (80.2)	458 (81.3)	0.797 (0.672, 0.945)
P. aeruginosa colonization at baseline: Positive	205 (35.7)	199 (35.3)	0.878 (0.700, 1.102)
P. aeruginosa colonization at baseline: Negative	370 (64.3)	364 (64.7)	0.772 (0.635, 0.940)
Baseline BSI score: <=4	150 (26.1)	148 (26.3)	0.686 (0.497, 0.947)
Baseline BSI score: 5-8	239 (41.6)	220 (39.1)	0.765 (0.604, 0.969)
Baseline BSI score: >=9	182 (31.7)	195 (34.6)	0.924 (0.728, 1.172)
Baseline BSI score: <Median	278 (48.3)	275 (48.8)	0.738 (0.589, 0.924)
Baseline BSI score: >=Median	293 (51.0)	288 (51.2)	0.856 (0.702, 1.044)
BE-CT score: <Median	273 (47.5)	255 (45.3)	0.910 (0.726, 1.141)
BE-CT score: >=Median	302 (52.5)	308 (54.7)	0.743 (0.609, 0.906)
Baseline FEV1 % predicted post-bronchodilator: <50%	102 (17.7)	98 (17.4)	1.227 (0.862, 1.747)
Baseline FEV1 % predicted post-bronchodilator: >=50%	469 (81.6)	465 (82.6)	0.718 (0.607, 0.849)
Stratification region: North America	83 (14.4)	81 (14.4)	0.830 (0.586, 1.176)
Stratification region: Europe	221 (38.4)	221 (39.3)	0.857 (0.681, 1.078)



Subgroup	Brensocatib 25mg QD (N=575) n (%)	Placebo (N=563) n (%)	Rate Ratio (95% CI)
Overall	575 (100.0)	563 (100.0)	0.806 (0.694, 0.936)
Stratification region: Japan	28 (4.9)	29 (5.2)	0.323 (0.140, 0.746)
Stratification region: Rest of World	243 (42.3)	232 (41.2)	0.827 (0.648, 1.056)
Geographic region: South America	157 (27.3)	157 (27.9)	0.920 (0.674, 1.256)
Geographic region: Eastern Europe	68 (11.8)	72 (12.8)	0.617 (0.352, 1.083)
Geographic region: Western Countries	205 (35.7)	201 (35.7)	0.857 (0.694, 1.057)
Geographic region: Asian Countries	91 (15.8)	88 (15.6)	0.622 (0.412, 0.941)
Geographic region: Oceania	54 (9.4)	45 (8.0)	0.772 (0.507, 1.177)
Baseline eosinophil count: >=300/mm3	111 (19.3)	106 (18.8)	0.863 (0.618, 1.203)
Baseline eosinophil count: <300/mm3	461 (80.2)	452 (80.3)	0.794 (0.671, 0.939)
Smoking status: Former smoker	163 (28.3)	183 (32.5)	0.746 (0.576, 0.966)
Smoking status: Never smoked	412 (71.7)	380 (67.5)	0.844 (0.703, 1.013)
Use of inhaled steroids: Yes	324 (56.3)	352 (62.5)	0.761 (0.628, 0.922)
Use of inhaled steroids: No	251 (43.7)	211 (37.5)	0.934 (0.729, 1.197)
History of asthma: Yes	109 (19.0)	111 (19.7)	0.986 (0.724, 1.342)
History of asthma: No	466 (81.0)	452 (80.3)	0.760 (0.641, 0.901)
History of COPD: Yes	83 (14.4)	102 (18.1)	0.808 (0.555, 1.175)
History of COPD: No	492 (85.6)	461 (81.9)	0.804 (0.683, 0.946)
Hospitalized in prior 24 months for PE: Yes	133 (23.1)	142 (25.2)	0.807 (0.608, 1.071)
Hospitalized in prior 24 months for PE: No	442 (76.9)	421 (74.8)	0.809 (0.679, 0.964)

Note: Protocol-defined PEs are based on the independent adjudication process.
BE-CT = Bronchiectasis-Computed Tomography, BSI = Bronchiectasis Severity Index, CI = confidence interval, COPD = chronic obstructive pulmonary disease, CT = computed tomography, ITT = intent-to-treat, FEV1 = forced expiratory volume in 1 second, NE = not estimable, PE = pulmonary exacerbation, QD = once daily.

In the FEV_1 <50% predicted subgroup (~100 patients/arm), a treatment-by-FEV_1 interaction test yielded p=0.095 (10 mg vs placebo) and 0.0053 (25 mg vs placebo), suggesting potential heterogeneity; however, both brensocatib 10 mg and 25 mg groups showed rate ratios <1 compared with placebo when stratified by FEV_1 above/below median.

Ancillary analyses

Several ancillary analyses were conducted to support the interpretation of primary and secondary efficacy results and to explore pharmacodynamic (PD) and patient-reported outcome (PRO) relationships.

Sensitivity analyses for the primary endpoint

Multiple sensitivity analyses were performed to test the robustness of the primary analysis assumptions (e.g. missing at random).

The *tipping point analyses*, which assessed treatment effect using multiple imputation under various missing not at random assumptions, is provided in the table below (25mg only).

Table 73: Sensitivity Analysis 1: Primary Endpoint, Annualized Rate of Pulmonary Exacerbations - Observed p-Values from the Tipping Point Analysis, Intent-to-Treat Analysis Set

	Brensocatib 25mg QD								
Placebo	0.2	0.6	1	1.4	1.8	2.2	2.6	3	3.4
0.2	0.004	0.011	0.029	0.070	0.126	0.233	0.378	0.540	0.741
0.6	0.001	0.004	0.012	0.031	0.063	0.132	0.232	0.355	0.526
1.0	<0.001	0.001	0.005	0.013	0.030	0.068	0.129	0.222	0.359
1.4	<0.001	<0.001	0.002	0.005	0.014	0.034	0.070	0.129	0.242
1.8	<0.001	<0.001	<0.001	0.002	0.007	0.018	0.042	0.067	0.146
2.2	<0.001	<0.001	<0.001	0.001	0.003	0.008	0.024	0.039	0.087
2.6	<0.001	<0.001	<0.001	<0.001	0.001	0.004	0.010	0.024	0.043
3.0	<0.001	<0.001	<0.001	<0.001	<0.001	0.002	0.005	0.012	0.029
3.4	<0.001	<0.001	<0.001	<0.001	<0.001	0.001	0.003	0.005	0.013

The *Jump-to-Reference analysis*, in which the placebo group acted as the reference after an ICE related to study treatment, is provided in the table below.

Table 74: Sensitivity Analysis 2: Primary Endpoint, Annualized Rate of Pulmonary Exacerbations - Reference-Based Multiple Imputation - On-Study Estimand, Intent-to-Treat Analysis Set

Characteristics	Brensocatib 10mg QD (N=583)	Brensocatib 25mg QD (N=575)	Placebo (N=563)
Annualized rate (95% CI)	1.024 (0.917, 1.145)	1.048 (0.937, 1.173)	1.287 (1.159, 1.429)
Rate ratio vs Placebo (95% CI)	0.796 (0.685, 0.925)	0.815 (0.700, 0.948)	
p-Value	0.0029	0.0079	

Post-hoc re-analysis of the primary endpoint using the original 28-day

The primary endpoint was re-analysed using the original 28-day and the amended 14-day separation windows, applying the same ITT set, on-study estimand, model and covariates. As expected, PE counts increased in all arms with the 14-day window, but differences vs placebo and rate ratios were essentially unchanged; brensocatib remained statistically significant and clinically meaningful under both windows. A side-by-side table and footnotes specify the negative binomial model and covariates used (Table 75).

Table 75: Study INS1007-301: Summary and Statistical Analysis of Primary Endpoint: Annualized Rate of Pulmonary Exacerbations. Two PEs separated by at least 28 days vs Two PEs separated by at least 14 days - On-Study Estimand (ITT Analysis Set)

	Window A 2 PEs separated by at least 28 days			**Window B 2 PEs separated by at least 14 days**		
	Brensocatib 10 mg QD [N=583]	**Brensocatib 25 mg QD [N=575]**	**Placebo [N=563]**	**Brensocatib 10 mg QD [N=583]**	**Brensocatib 25 mg QD [N=575]**	**Placebo [N=563]**
Participants with ≥1 exacerbation event, n (%)	292 (50.1)	288 (50.1)	324 (57.5)	292 (50.1)	288 (50.1)	324 (57.5)
Number of participants with exacerbation events, n (%)						
0	291 (49.9)	287 (49.9)	239 (42.5)	291 (49.9)	287 (49.9)	239 (42.5)
1	161 (27.6)	155 (27.0)	153 (27.2)	153 (26.2)	146 (25.4)	145 (25.8)
2	66 (11.3)	70 (12.2)	90 (16.0)	63 (10.8)	67 (11.7)	88 (15.6)
≥3	65 (11.1)	63 (11.0)	81 (14.4)	76 (13.0)	75 (13.0)	91 (16.2)
Total number of exacerbation events	520	508	617	563	554	669
Total time at risk in patient-years [1]	531.98	523.71	506.31	534.30	526.22	509.02
Annualized rate (95% CI) [2]	0.946 (0.851, 1.052)	0.953 (0.857, 1.060)	1.190 (1.077, 1.316)	1.015 (0.910, 1.132)	1.036 (0.927, 1.157)	1.286 (1.158, 1.428)
Rate ratio vs Placebo (95% CI) [2]	0.795 (0.688, 0.917)	0.801 (0.694, 0.924)		0.789 (0.680, 0.916)	0.806 (0.694, 0.936)	
p-Value [2]	0.0017	0.0023		0.0019	0.0046	
Difference (14-day vs 28-day)						
Rate Absolute (% change)	0.069 (+7.3)	0.083 (+8.7)	0.096 (+8.0)			
Rate Ratio (% change)	0.006 (+0.75)	0.005 (+0.62)				

Cut-off date 28 March 2024.

Window A: A minimum of 4 weeks (28 days) must occur between the end date of an earlier PE and the start date of the next PE, otherwise, both PEs will be considered the same exacerbation.

Window B: A minimum of 2 weeks (14 days) must occur between the end date of an earlier PE and the start date of the next PE, otherwise, both PEs will be considered the same exacerbation.

Time at risk is calculated as: date of Week 52 or date of early study discontinuation - date of randomization + 1 minus the sum of number of days the participant experiences PE events that meet the protocol definition of PE diagnosis (end date of the PE - start date of the PE + 1).

[1] The total time at risk of person-years is calculated by pooling all participants time at risk in years, separately for each treatment.

[2] Analysis of total number of exacerbation events is based on a negative binomial model including treatment group, sputum sample being classified as positive or negative for Pseudomonas aeruginosa at screening visit, the number of prior pulmonary exacerbations [<3 or >=3] in the previous 12 months, and stratification region (North America, Europe, Japan, and the Rest of World) and age group (adult, adolescent). The robust (empirical) sandwich estimator is used for the standard errors of the rates and rate ratios.

[3] PE events with no end date at Week 52 visit, the date of Week 52 visit is used as end date for the particular PE. The time at risk (natural log years) is the offset.

CI = confidence interval; ITT = intent-to-treat; PE = pulmonary exacerbation; QD = once a day.

Patient-reported outcome (PRO) anchor-based analysis

RTI-HS Project No. 0307201: Meaningful Within-Patient Change Assessment of the Respiratory Symptoms Domain of the Quality of Life Questionnaire–Bronchiectasis (ASPEN data)

A psychometric evaluation of the QOL-B Respiratory Symptoms domain score was conducted using data from the ASPEN study (INS1007-301) to establish a meaningful within-patient change (MWPC) threshold. The QOL-B instrument was administered biweekly from baseline to Week 56.

The primary anchor for determining clinical relevance was the Patient Global Impression of Severity (PGIS); the Patient Global Impression of Change (PGIC) served as a secondary anchor. The analysis included descriptive statistics, test-retest reliability (ICC = 0.88 for stable patients), and anchor-based as well as distribution-based methods.

The resulting MWPC threshold for improvement in the Respiratory Symptoms domain score was estimated to be in the range of 11–14 points, consistent with the QOL-B item structure (each item contributes in 3.7-point increments). These values are supported by:

- Mean and median change scores in patients with 1-point improvement on the PGIS (12.6 and 13.0),
- Concordant estimates from patients rated as "much improved" on the PGIC (mean 10.0, median 11.1),
- And distribution-based thresholds (half-SD: 8.5; SEM: 5.8).

6.3.3. Supportive studies

Study INS1007-201 (WILLOW)

Study design:

INS1007-201 was a Phase 2, randomized, double-blind, placebo-controlled, parallel-group, multicenter, multinational study evaluating the efficacy, safety, PK, and PD of brensocatib in adults with NCFBE.

Objectives and endpoints:

Primary endpoint: Time to first protocol-defined pulmonary exacerbation (PE) over 24 weeks.

Key secondary endpoints: Annualized rate of PEs, change from baseline in post-bronchodilator FEV1, and change in Quality of Life Questionnaire-Bronchiectasis (QOL-B) Respiratory Symptoms Domain Score.

Population:

A total of 256 participants were randomized (1:1:1) to receive brensocatib 10 mg, 25 mg, or placebo once daily for 24 weeks. Participants were adults aged 18-85 years with HRCT-confirmed NCFBE and ≥2 documented PEs requiring antibiotics in the previous year.

Results:

Primary Endpoint Analysis - Time to first PE:

Table 76: Kaplan Meier and Stratified Log Rank Analysis of Time to First Pulmonary Exacerbation (ITT Population)

	Placebo (N=87)	INS1007 10 mg (N=82)	INS1007 25 mg (N=87)
Subject Status:			
At least one exacerbation, n (%)	42 (48.3)	26 (31.7)	29 (33.3)
Censored, n (%)	45 (51.7)	56 (68.3)	58 (66.7)
Stratified[a] Log-rank test *P*-value[b]	--	0.014	0.022
Un-stratified Log-rank test *P*-value[b]	--	0.010	0.028
Kaplan-Meier Estimates (days):	--	--	--
Median	189.0	NE	NE
90% CI of Median	(141.0, NE)	(NE, NE)	(NE, NE)
80% CI of Median	(152.0, NE)	(NE, NE)	(NE, NE)
Q1, Q3	(67.0, NE)	(134.0, NE)	(96.0, NE)
Min, Max[c]	(2, 204)	(7, 276)	(3, 211)

-

[a] Stratified by P aeruginosa colonization status and maintenance antibiotic use at baseline

[b] P values are one-sided for superiority

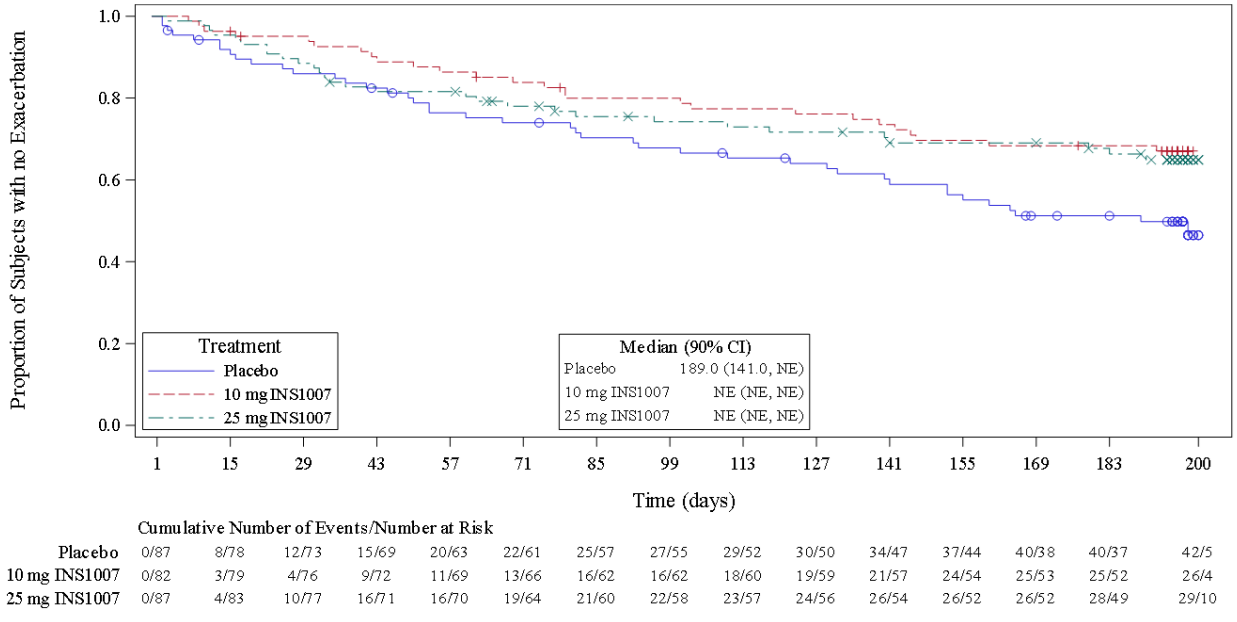
[c] Maximum value is censored

Notes: Time to first exacerbation defined as the time (days) from the date of randomization to the date of first documentation of exacerbation. Percentages are based on the number of subjects in the ITT Population.

CI = confidence interval; ITT = intent-to-treat; Max = maximum; Min = minimum; NE = not evaluable; Q1 = first quartile; Q3 = third quartile

Forty-two (42) subjects (48.3%) in the placebo group experienced an exacerbation compared to 26 subjects (31.7%) and 29 subjects (33.3%) in the 10 mg and 25 mg groups, respectively (Cochran-Mantel-Haenszel test P value = 0.017 and 0.019 for 10 mg and 25 mg versus placebo, respectively). The median time to first exacerbation in the placebo group was 189 days, while the median time to first exacerbation could not be estimated for either INS1007 group due to the low number of exacerbations in each group. The stratified log-rank test one-sided P values for time to first exacerbation compared to placebo were P = 0.014 and P = 0.022 for the 10 mg and 25 mg groups, respectively. The equivalent two-sided P values were P = 0.027 for the 10 mg group and P = 0.044 for the 25 mg group (See figure below).

Figure 36: Kaplan Meier Plot of Time to First Pulmonary Exacerbation (ITT Population)



Source: CSR - Figure 14.2.1.1.1.3.-
Notes: Time to first exacerbation = time (days) from date of randomization to date of first documentation of exacerbation. Censored values are indicated on the graph.
NE=not estimable

Sensitivity Analyses - Proportional Hazard Assumption and Cox Proportional Hazard Ratio

The proportional hazard assumptions for the Cox model were tested and validated. There was a statistically significant treatment effect for both INS1007 10 mg and 25 mg, on time to pulmonary exacerbation compared with placebo, consistent with the results of the primary analysis. The stratified hazard ratio for INS1007 10 mg vs. placebo was 0.578 (90% CI 0.382, 0.875, one-sided P = 0.015).

The stratified hazard ratio for the 25 mg dose was 0.615 (90% CI 0.412, 0.918, one-sided P = 0.023). The equivalent two-sided P values were 0.029 and 0.046 for the 10 mg and 25 mg doses, respectively.

Secondary Analyses of Primary Endpoint - Annualized Rate of Pulmonary Exacerbations Over the 24-Week Treatment Period

The unadjusted rate of pulmonary exacerbations per person-year was 0.80 and 0.96 in the

INS1007 10 mg and 25 mg groups, respectively, compared with 1.25 in the placebo group. Treatment group differences were statistically significant for the INS1007 10 mg dose compared with placebo (RR = 0.64, 95% CI = 0.42, 0.99; two-sided P =0.043) and did not reach statistical significance for the INS1007 25 mg dose (RR = 0.77, 95% CI = 0.52, 1.15; two-sided P = 0.205).

Sensitivity Analyses for Rate of Pulmonary Exacerbation

- Adjusted Annualized Rate: The annualized rate of pulmonary exacerbations was statistically adjusted for treatment group, baseline strata and treatment duration effects. The INS1007 10 mg treatment effect was statistically significant for the adjusted rate (RR = 0.64, 95% CI = 0.42, 0.98; two-sided P = 0.041); the INS1007 25 mg treatment effect did not reach statistical significance (RR = 0.752, 95% CI = 0.50, 1.13; two-sided P = 0.167).

- Time to Recurrent Event Analysis (Anderson-Gill Model): Using the unstratified Hazard Ratio, the analysis showed a statistically significantly longer time to recurrent pulmonary exacerbation events in the INS1007 10 mg group compared with placebo: Unstratified HR=0.648, two-sided P = 0.047. The unstratified hazard ratio in the INS1007 25 mg group was 0.768 (two-sided P = 0.201). An ad hoc analysis of 95% CI's around the hazard ratios showed the 10 mg dose maintaining superiority in the unstratified analysis (95% CI = 0.422, 0.995).

Secondary Endpoint Analyses

- Change From Baseline in QoL-B Respiratory Symptoms Score (MMRM): Mean Baseline scores for the QoL-B Respiratory Symptoms Score were 55.2± 19.2 and 56.1±18.9 in the INS1007 10 mg and 25 mg groups, respectively, compared with 51.9±18.7 in the placebo group. LS mean changes (SE) from Baseline to the Last Assessment Visit were 3.8 (0.78) and 5.9 (0.76) in the INS1007 10 mg and 25 mg groups compared with 5.7 (0.77) in the placebo group. The LS mean (SE) difference from placebo was −2.0 (1.01) for INS1007 10 mg and 0.2 (1.01) for INS1007 25 mg; the differences were not statistically significant. There were no statistically significant differences between active treatment groups and placebo when fixed effects of treatment, P. aeruginosa colonization status, or Baseline maintenance antibiotic use were added to the model.

- Change From Screening in Post-bronchodilator ppFEV1: Analysis of covariance (ANCOVA) of the changes from Screening in post-bronchodilator ppFEV1 over the 24-week Treatment Period at Weeks 12 and 24 showed no statistically significant differences among the treatment groups.

6.3.4. Overall discussion and conclusions on clinical efficacy

6.3.4.1. Discussion

Product Development and Regulatory Interactions

Brensocatib is an oral, immediate-release, film-coated tablet developed in 10 mg and 25 mg strengths. The proposed commercial tablet is consistent with that used in the pivotal Phase 3 ASPEN study, with the only difference being the colour of the film coating. The 25 mg dose is the applicant's proposed therapeutic dose for routine clinical use. Brensocatib has received PRIME designation from EMA, reflecting its potential to address an unmet medical need. The development programme has benefited from continuous interaction with CHMP / SAWP, including scientific advice and protocol assistance. These interactions addressed critical aspects of the clinical programme, such as study design, statistical methodology, dose selection, inclusion of adolescents, and the overall pharmacology strategy.

Study Design and Population

The pivotal ASPEN study, together with the supportive Phase 2 WILLOW study, forms the basis for the assessment of brensocatib's efficacy and safety. ASPEN was a global, randomized, double-blind, placebo-controlled trial evaluating the efficacy and safety of brensocatib in patients with NCFBE and ⩾ 2 prior pulmonary exacerbations (⩾1 in adolescents) requiring systemic antibiotics within the last 12 months.

The study was designed to randomize approximately 1620 adults in a 1:1:1 ratio to receive brensocatib 10 mg QD, brensocatib 25 mg QD, or placebo QD for 52 weeks (~540 participants per arm). The study was designed to randomize approximately 40 adolescents ⩾12 to <18 years of age in a 2:2:1 ratio to receive brensocatib 10 mg QD, brensocatib 25 mg QD, or matching placebo QD for 52 weeks (16:16:8 participants per arm, respectively). There was no stratification for adolescents. Sputum samples were classified as positive or negative for P. aeruginosa at Screening for all adult participants. Adolescent

participants were not required to produce a sputum sample at any time during the study if they were unable. The study consisted of a Screening Period of up to 6 weeks, a Treatment period of 52 weeks, and an End of Study Visit 4 weeks after the end of treatment.

Eligibility criteria is considered adequate to characterise broad demographic profiles of NCFB, with some notable distinctions between the ASPEN study population and general disease characteristics with respect to aetiology, endotype representation, and clinical phenotype. Multiple pathogenic endotypes may coexist across different underlying causes, including a type 2 inflammatory endotype in patients with concomitant asthma and other conditions. Patients with known immunodeficiency or receiving immunomodulatory therapy were excluded, thereby minimizing the inclusion of bronchiectasis cases driven by immune dysfunction.

Beyond aetiology and endotype, phenotypic stratification is also clinically relevant. Frequent exacerbator phenotypes are often defined by ⩾3 exacerbations per year and associated with increased risk of future exacerbations, hospitalizations, and mortality. Inclusion criteria required participants to have experienced ⩾2 exacerbations in the 12 months prior to screening (⩾1 for adolescents). Randomization was managed to ensure balanced representation across these key subgroups. Approximately 30% of adult participants should not have had a history of ⩾3 pulmonary exacerbations (PEs) in the prior year. Enrolment was capped such that no more than 20% of participants were older than 75 years, <20% with peripheral blood eosinophil count ⩾300 cells/mm^3 at screening, or <20% with comorbid chronic obstructive pulmonary disease (COPD). This is endorsed since these are usually handled differently in clinical practice and could have confounded treatment implications.

While the key inclusion criterion was based on PE history, it was clarified that investigators were instructed via case report form completion guidelines (CCGs) and training to verify prior exacerbations using documented evidence of physician-prescribed antibiotics. The applicant' s documentation adequately operationalized the enrichment criterion ("⩾2 pulmonary exacerbations in the prior 12 months defined by need for antibiotic prescription") across screening, data capture, and monitoring. The Indication History page in the electronic data capture (EDC) was 100% source data verified by clinical research associates (CRAs), who confirmed eligibility against inclusion/exclusion criteria, including review of prescription records and country-specific requirements. Nevertheless, the absence of explicit instructions in the CSP was considered a limitation. Despite this, the study population reflected the intended treatment population. Therefore, this issue was not further pursued.

The applicant also presented an appropriate and standard set of minimum washout periods for bronchodilators and related medications before spirometry testing. These intervals are consistent with international guidelines (e.g., ATS/ERS) to ensure that appropriate pulmonary function tests reflect baseline lung function rather than temporary medication effects.

A total of 2,296 participants were screened, of whom 529 (23.0%) failed screening, primarily due to not meeting inclusion/exclusion criteria (20.6%). 1,767 participants (77.0%) were randomized across three arms: 595 (25.9%) to 10 mg IP, 593 (25.8%) to 25 mg IP, and 579 (25.2%) to placebo. After accounting for re-screened participants, 1,721 randomized participants were analysed (583 in 10 mg, 575 in 25 mg, and 563 in placebo groups). The study population comprised of predominantly adults (97.6%), with 41 adolescents enrolled (2.4%), which is acceptable.

Study completion rates were approximately 80% overall, with 78.6%, 81.0%, and 81.2% completing the study in the 10 mg, 25 mg, and placebo groups, respectively. Discontinuations prior to study completion occurred in 12.7% of participants, primarily due to subject withdrawal (6.3%), adverse events (1.7%), and death (0.8%). Discontinuation rates of the IP were slightly lower, with 86.7% completing IP administration and 12.0% discontinuing IP prematurely, mainly due to adverse events (4.1%) and subject withdrawal (5.2%) as the highest rate of discontinuations, similarly between arms. Lost to follow-up accounted for less than 1% of discontinuations. These rates are not considered to

have impacted the results. Of note, a small proportion of participants discontinued due to external factors such as the war in Ukraine (1.9% overall).

The study underwent multiple protocol amendments throughout its conduct, which coincide with adaptations for emerging operational requirements and input from scientific advice. The amendments were implemented prospectively, before database lock and unblinding. Timing and content of the changes include adjustments to endpoint definitions, inclusion/exclusion criteria, multiplicity control and incorporation of covariates (age for including adolescents in the trial and including them in the primary analysis). Changes in enrolment targets aimed at managing recruitment and demographic representation could introduce complexities if randomisation scheme was altered, however, this appears not to be the case since the number of randomised participants per month did not differ substantially from rates of enrolment previous to the implemented change.

Prior to database lock and unblinding, major protocol deviations were identified. These deviations were balanced across treatment groups, with 8.2%, 9.6%, and 7.6% of participants in the 10 mg, 25 mg, and placebo groups, respectively, having at least one major deviation. The most common deviation was related to study treatment administration or dispensation, occurring in 3.1%, 3.5%, and 4.3% of participants across respective groups, primarily due to treatment compliance outside the 75–120% range. Notably, two participants in the 10 mg group received incorrect treatments at certain visits. Other deviations included issues with concomitant medication use (1.6–2.3%), exclusion (1.6–2.4%) and inclusion criteria (0.2–1.4%), and study procedures or assessments (<0.5%). Given the balanced distribution of deviations across groups, low overall frequency, and lack of any discernible pattern, these major protocol deviations are deemed unlikely to have had any meaningful impact on the study's overall efficacy outcomes.

While most baseline variables are well balanced, Pseudomonas status and FEV1 presented a moderate risk of confounding for interpreting efficacy. This was accounted for in the adjusted analyses for Pseudomonas status. Overall, participants had comparable lung function at baseline, with mean FEV_1 % predicted ranging from 72–74%, indicating mild to moderate impairment. About 17.5% had FEV1 <50%, majority (~82%) had >50%, so the study cohort is largely moderately impaired. Post-bronchodilator spirometry patterns had a heterogeneous distribution of lung function. About 50% of participants had an obstructive pattern (FEV_1/FVC <0.7), characteristic of COPD or bronchiectasis with airflow limitation. PRISm phenotype (FEV_1 <80% with preserved FEV_1/FVC $\geqslant$0.7) was present in 18.5%, suggestive of restrictive or non-obstructive impairment. The remaining 31% had normal lung function. Overall, this distribution reflects the expected heterogeneity in pulmonary physiology typical of the broader NCFB population.

COPD history was higher (18.1%) in placebo than in the IP arms (13–14%). However, since inhaled corticosteroid use was higher (62.5%) than in the IP arms (55–56%) this is expected. Therefore it can be concluded that there was no apparent bias between arms to inflate brensocatib treatment effect.

Primary Endpoint and Adjudication Process

The primary efficacy endpoint was the annualized rate of pulmonary exacerbations (PEs), defined by worsening of $\geqslant$3 respiratory symptoms for $\geqslant$48 hours, requiring systemic antibiotic treatment. Events were adjudicated by an independent Clinical Endpoint Committee (CEC), ensuring objective, protocol-consistent classification. The definition is consistent with ERS guidelines and previous bronchiectasis trials. Pulmonary exacerbations were considered as severe if requiring treatment with intravenous antibiotics and/or resulted in hospitalisation.

Severe PEs were defined as those requiring intravenous antibiotics and/or hospitalization $\geqslant$24 hours, in accordance with standard clinical severity grading. In the original protocol, a new PE was to be distinguished from a prior episode if $\geqslant$28 days had elapsed between the end of one event and the start

of the next one. However, following blinded review and EMA scientific advice, Protocol Amendment 4 (13 February 2024) introduced a 14-day interval, which was applied retrospectively to all 2,267 adjudicated events. This reclassification resulted in 57 events being split into 64, increasing the total to 2,331 PEs (53 of which were protocol-defined). Upon CHMP request, the applicant provided a re-analysis showing stable treatment effects with only minor, non-relevant changes in rates and rate ratios. Brensocatib remained statistically significant and clinically meaningful under both windows. The CEC re-review and retrospective application of the 14-day window are transparently reported.

A monthly dossier of source documents was submitted to the CEC, with each adjudication based solely on materials specific to the event in question. The CEC process involved independent dual review, with discordant cases escalated to full committee review (54 meetings held). All 2,267 investigator-reported events were adjudicated, and of these, 1,897 (83.7%) met the protocol-defined PE criteria, while 370 (16.3%) did not. Of the non-protocol-defined events, 212 (57.3%) were considered Clinically Relevant Exacerbations (CREs). The primary efficacy analyses included only protocol-defined, CEC-adjudicated PEs, while CREs were assessed in a pre-specified supplementary analysis. From a clinical perspective, this is a well-structured approach to PE definition. Adjudication is appropriate and necessary given the heterogeneity of disease and variability in clinical presentation.

Statistical Considerations

The primary analysis utilized a treatment policy estimand to evaluate the annualized rate of pulmonary exacerbations (PEs), incorporating all observed PE events regardless of intercurrent events (ICEs), such as treatment discontinuation or use of prohibited medications. This approach aligns with the ICH E9(R1) addendum and reflects ITT perspective. To assess robustness, a supplementary analysis employed an alternative treatment policy estimand with a uniform time-at-risk, standardizing exposure duration across participants by fixing the risk period to the total time on study. This duration-standardized annualized PE rate served to validate assumptions about patient follow-up and event rates.

Sensitivity analyses included a reference-based "jump-to-reference" (J2R) approach. The applicant clarified that observed data after ICEs were retained, and only genuinely missing data were imputed using placebo-arm distributions, consistent with the treatment policy strategy and the pre-specified SAP. Additional "while on treatment" estimands, censoring events after treatment discontinuation or other ICEs, were also conducted.

A multiplicity adjustment strategy was used, with an enhanced mixture-based gatekeeping procedure, incorporating a truncated Hochberg test for the primary endpoint family (Family 1) at a two-sided alpha level of 0.01 (truncation fraction = 0.9). This design allowed for flexible alpha allocation, preserving most of the type I error rate for the primary endpoint while permitting the full family-wise alpha of 0.05 for testing secondary endpoints only if both doses met the primary endpoint.

Missing data were handled under the MAR assumption, which may not hold in this population. The assumption that PE rates post-discontinuation are similar to on-treatment periods is implausible. Accordingly, MNAR sensitivity analyses were conducted, and while the extent of missing data was moderate, such analyses remain important for interpreting the treatment effect. In response to the CHMP request, the applicant provided a reference-based "jump-to-reference" (J2R) analysis for both the primary endpoint and the FEV_1 secondary endpoint. The applicant clarified that observed data after intercurrent events were retained, and only genuinely missing data were imputed using placebo-arm distributions, which is consistent with a treatment policy strategy and the SAP. This clarification addressed earlier concerns regarding inappropriate replacement of observed data post-ICE.

The gatekeeping strategy (enhanced mixture-based) ensured family-wise error control across key endpoints. However, the absence of informative confidence intervals aligned with the hierarchy limits interpretation. Changes to the statistical methods (including multiplicity procedures, additional

sensitivity analyses, and the shortened 14-day PE separation rule) were all implemented prospectively, prior to database lock and unblinding.

Both the 10 mg and 25 mg doses met the primary endpoint in the primary analysis (cut-off 28-Mar-2024), with adjusted p-values of 0.0038 and 0.0048, respectively. Each dose demonstrated statistically significant and reductions in annualized rate of pulmonary exacerbations (PEs) compared to placebo across multiple estimands (On-IP, On-Treatment, and On-Study). Rate ratios ranged from approximately 0.78 to 0.82, with nominal p-values consistently <0.01.

The supplementary analysis of CREs (events requiring antibiotics but not meeting protocol PE criteria) confirmed no treatment effect (annualised CRE rate: 0.128 [10 mg], 0.111 [25 mg], 0.133 [placebo]; rate ratios 0.97 and 0.83, respectively), consistent with the primary endpoint.

The adjudication process, including the involvement of a CEC, strengthens the reliability of this symptom-based primary endpoint. However, the retrospective application of the 14-day rule to already adjudicated events is considered a critical methodological concern. A side-by-side analysis using both the original 28-day and amended 14-day windows was also requested to further evaluate the potential impact on the primary endpoint results. Both 28 day and 14 days results showed a positive outcome.

Secondary Endpoints

As both doses met the gatekeeping criteria, secondary endpoints were tested using the full two-sided alpha of 0.05, with adjustments for multiplicity within each family.

Time to First Pulmonary Exacerbation

Both doses achieved statistical significance for Time to first PE (adjusted p = 0.0200 for 10 mg; 0.0364 for 25 mg). The median delay was approximately 13 weeks, reflecting a sustained treatment effect and supporting the primary endpoint findings.

Exacerbation-Free Status

Both doses achieved statistical significance for Responder status for exacerbation-free participants at Week 52 (adjusted p = 0.0200 for 10 mg; 0.0364 for 25 mg). The proportion of patients who remained exacerbation-free throughout the 52-week study period was higher in the brensocatib arms (48.5%) compared to placebo (40.3%). This binary responder outcome, based on adjudicated events, complements the primary endpoint and reflects clinically relevant individual-level efficacy.

Post-Bronchodilator FEV_1

Lung function was assessed via change in post-bronchodilator FEV_1. In line with the CHMP request, an additional J2R analysis based on the full ITT on-study estimand has been provided. Results confirmed no statistically significant effect for brensocatib 10 mg (LS mean difference vs placebo +0.010 L; 95% CI −0.017 to 0.037; p=0.4793) and a small but statistically significant effect for brensocatib 25 mg (LS mean difference vs placebo +0.036 L; 95% CI 0.008 to 0.064; p=0.0120). While this supports the robustness of the findings to assumptions about missing data, the absolute magnitude of change remains modest (~36 mL benefit for 25 mg vs placebo) and the clinical relevance is unclear, as no bronchiectasis-specific MCID for FEV_1 has been established.

Annualized Rate of Severe Exacerbations

Brensocatib 10 mg and 25 mg both showed a numerical reduction in severe PE rates (0.137 events/year) compared with placebo (0.185 events/year), corresponding to ~26% rate reductions. However, these differences were not statistically significant (nominal p-values of 0.1277 and 0.1025, respectively). Despite the lack of significance, the direction and consistency of the effect are noted.

Quality of Life – QOL-B Respiratory Symptoms Domain

The QOL-B Respiratory Symptoms Domain score showed numerical improvement in both brensocatib arms. LS mean differences from placebo at Week 52 were 2.031 points (10 mg, nominal p = 0.0594) and 3.766 points (25 mg, nominal p = 0.0004). However, as hierarchical testing was broken, the nominal p-value for 25 mg is not alpha-controlled and must be interpreted with caution. The applicant responses to CHMP request clarified that the eDiary system used for QOL-B data collection was validated, with direct data capture into the EDC. Twenty adult participants completed an unapproved US English version of the questionnaire due to a system error in certain countries; these data were excluded from the analysis. QOL-B was administered only to adults, as no validated adolescent translation was available. Importantly, the observed group-level changes did not reach the established meaningful within-patient change (MWPC) threshold of 11–14 points, limiting interpretation of a clinically meaningful improvement.

Determination of the minimal clinically important difference (MCID) for the QOL-B respiratory symptoms domain

A combination of anchor-based and distribution-based methods was used to estimate the minimal within-patient change (MWPC) in the QOL-B Respiratory Symptoms domain. The primary anchor-based estimate was derived from the mean (12.6) and median (13.0) change scores observed in patients reporting a 1-point improvement on the Patient Global Impression of Severity (PGIS) scale at Week 52. Supporting estimates from the "much improved" Patient Global Impression of Change (PGIC) group and distribution-based methods (half-standard deviation and standard error of measurement) further supported this threshold. Given the QOL-B scoring structure, where changes occur in 3.7-point increments, a threshold range of 11 to 14 was proposed, corresponding to 3–4 item-category improvements. This range exceeds measurement error and reflects improvement aligning with empirical data and prior literature. Worsening thresholds were not proposed due to sample size limitations and statistical constraints, which is considered appropriate.

At Week 52, participants with missing follow-up data were treated as non-responders. Using the 14-point MWPC threshold, the 25 mg arm showed a nominal benefit (OR 1.30; 95% CI 1.02–1.87; p = 0.038), but as hierarchical testing was not preserved, these results should be considered descriptive. With the lower 11-point threshold, responder rates were higher overall and similar between arms (35.5% for 10 mg, 33.8% for 25 mg, and 30.4% for placebo), with no statistically significant differences versus placebo. Overall, while there is directional consistency, the absence of alpha-controlled statistical significance and failure to reach the proposed MWPC limit the strength of conclusions regarding clinically meaningful benefit.

Subgroup Analyses

In subgroup analyses both the 10 mg and 25 mg doses demonstrated clinically meaningful and statistically significant reductions in pulmonary exacerbation (PE) rates compared to placebo, with overall similar efficacy. The 25 mg dose yielded a rate ratio (RR) of 0.806 (95% CI: 0.694 – 0.936), while the 10 mg dose achieved a comparable RR of 0.789 (95% CI: 0.680 – 0.916). Both doses reduced the proportion of patients experiencing ⩾1 exacerbation (50.1% for both brensocatib arms vs. 57.5% for placebo) and extended median time to first PE (49.0 and 50.7 weeks for 10 mg and 25 mg, respectively, vs. 36.7 weeks for placebo), with hazard ratios of 0.813 and 0.825, respectively. Responder analyses also supported consistent benefit for both doses, with odds ratios of 1.412 (10 mg) and 1.400 (25 mg) vs. placebo, with directional benefit preserved across most subgroups.

Important subgroups to highlight include women, patients with lower bronchiectasis severity index (BSI) scores, FEV1 ⩾50% and concurrent inhaled corticosteroid (ICS) use which exhibit greater magnitude of response with 25 mg versus placebo. Regional differences were notable, with higher efficacy in Asian and Japanese populations (RRs 0.622 and 0.323, respectively). However, the D90 responses confirmed that a formal treatment-by-FEV_1 interaction was not pre-specified in the SAP; post

hoc testing yielded p = 0.0950 for 10 mg vs placebo and p = 0.0053 for 25 mg vs placebo. The model for the <50% FEV_1 subgroup converged appropriately, using a sandwich covariance estimator, as did other similarly sized subgroups (e.g. baseline macrolide use, Asian region). Within the ⩽50% subgroup, baseline disease severity was higher (greater P. aeruginosa positivity, higher ICS and chronic macrolide use, higher BSI scores) compared with the ⩾50% subgroup, as expected. Although the 25 mg arm showed attenuation of FEV_1 decline and improvement in QOL-B RSS in the ⩽50% subgroup, no clear treatment effect on PE rate was observed. Given the small sample size (~100 per arm) and the exploratory, post hoc nature of these analyses, the results should be interpreted with caution.

Subgroups with baseline eosinophilia ⩾300 cells/mm^3, history of asthma, or FEV_1 <50% showed no evidence of treatment effect on the primary endpoint, consistent with possible reduced efficacy in populations with type 2 inflammatory features or advanced obstruction. Conversely, patients without P. aeruginosa colonisation (RR: 0.772) and those with FEV_1 ⩾50% predicted appeared to derive greater benefit. Numerically favourable but non-significant effects were observed in patients aged ⩾75 years and those with COPD or prior PE-related hospitalisations.

Overall, these subgroup findings indicate potential heterogeneity of treatment effect, with a statistically significant interaction observed post hoc for the 25 mg group by baseline FEV_1 (p = 0.0053). However, due to limited numbers and exploratory nature, the results are not considered definitive. The possibility of convergence issues in small subgroups was addressed by the applicant, who confirmed that models converged and that a sandwich covariance estimator was used (with parametric alternatives available if required). Subgroup results are therefore considered exploratory and should be interpreted cautiously.

Adolescents and Extrapolation

The adolescent subgroup (aged 12–<18 years) was too small for standalone efficacy conclusions. A Bayesian analysis was presented to support this, but CHMP noted that insufficient exploration of its operating characteristics (e.g. effective sample size by mixing weight) prevents robust conclusions. However, the extrapolation of efficacy to adolescents is supported by comparable pharmacokinetics similarility of the disease between both adult and adolescent populations. Therefore, for adolescents, section 5.1 of the SmPC clarifies that trends towards fewer pulmonary exacerbations and positive changes in post-bronchodilator FEV_1 were observed with 25 mg brensocatib versus placebo**.** Of note adolescents included had ⩾1 exacerbations compred to ⩾2 in the adult population. Nevertheless, it was agreed that the indication in patients with ⩾2 exacerbations would be acceptable for the adolescent population.

Supportive study

The WILLOW study provides supportive evidence over a 24-week period. The patient population was similar to the one in the pivotal ASPEN study, and representative of the target population, but did not enroll adolescents. Efficacy endpoints were similar but measured over shorter period of time (24 vs 52 weeks in the pivotal study). PEs had a minimum of 4 weeks required between onset. The treatment effect was shown for both doses (25 and 10 mg) of brensocatib in comparison to placebo with a statistically significant result for the primary endpoint, the median time to first exacerbation.

Justification of the 25 mg Dose

Both brensocatib 10 mg and 25 mg doses demonstrated a similar effect in reducing pulmonary exacerbation (PE) rates compared with placebo in the pivotal ASPEN study. The applicant has nevertheless proposed brensocatib 25 mg once daily as the recommended dose. This selection is based on the observation that the 25 mg dose demonstrated a consistently greater magnitude of effect across additional endpoints beyond exacerbation frequency, including lung function, patient-reported outcomes, and biomarker responses.

With respect to lung function, the 25 mg arm showed a smaller decline in post-bronchodilator FEV_1 at Week 52 (−24 mL) compared with placebo (−62 mL) and with the 10 mg arm (−50 mL). The J2R sensitivity analysis confirmed a modest but statistically significant effect for 25 mg versus placebo (LS mean difference +0.036 L; 95% CI 0.008–0.064; nominal p = 0.0120), whereas no significant difference was observed for 10 mg (+0.010 L; 95% CI −0.017 to 0.037; p = 0.4793). Although the absolute magnitude of the effect (~36 mL) is small and its clinical relevance in NCFBE remains uncertain (no established MCID), preservation of lung function is considered clinically important given its recognised association with morbidity and mortality in bronchiectasis. The applicant emphasised that the 25 mg group demonstrated an approximate 61% reduction in FEV_1 decline compared with placebo, bringing the rate of decline closer to that observed in healthy individuals.

The 25 mg dose also showed a larger attenuation of post-bronchodilator FVC decline (−75 mL vs placebo; nominal p <0.0001), and nominally significant improvements in QOL-B Respiratory Symptoms (RSS) and BEST scores, although these analyses were exploratory and not alpha-controlled. Furthermore, pharmacodynamic data indicated greater reductions in neutrophil serine protease (NSP) activity with 25 mg (NE −35.9%, CatG −58.0%, PR3 −46.9%) compared with 10 mg (NE −29.3%, CatG −36.0%, PR3 −23.6%). Exposure–response analyses demonstrated that nearly all patients receiving 25 mg achieved AUC_T values above the identified threshold for reduced FEV_1 decline (99.4% vs 44.5% in the 10 mg group).

Taken together, these findings suggest that the 25 mg dose provides broader and more consistent benefits than 10 mg, particularly in terms of lung function preservation and biomarker suppression, while maintaining an acceptable safety profile. The CHMP considered the applicant's justification for selecting the 25 mg dose as appropriate, though noted that the absolute size of the observed FEV_1 benefit is modest and its direct clinical relevance remains uncertain in NCFBE. Nonetheless, the totality of evidence was deemed to support 25 mg as the recommended dose for the proposed indication.

6.3.4.2. Conclusions on the clinical efficacy

Brinsupri (brensocatib) is intended for treatment of non-cystic fibrosis bronchiectasis (NCFBE) in patients 12 years of age and older with ≥2 prior pulmonary exacerbations requiring systemic antibiotics within the last 12 months. Submission is based on one Phase 3 pivotal study (INS1007-301, ASPEN) and a Phase 2 supportive study (INS1007-201).

The pivotal ASPEN study demonstrated that brensocatib, at both 10 mg and 25 mg doses, significantly reduced the annualized rate of pulmonary exacerbations in patients with NCFBE compared with placebo. These findings were supported by improvements in key secondary endpoints, including prolonged time to first exacerbation and a higher proportion of exacerbation-free participants over 52 weeks. The results were consistent across sensitivity analyses and adjudicated endpoints, lending robustness to the efficacy conclusions.

Additional endpoints showed modest effects: the 25 mg dose achieved a small but statistically significant attenuation of post-bronchodilator FEV_1 decline (+36 mL vs placebo; nominal p=0.0120), with consistent directional trends in FVC, QoL (QOL-B RSS), and biomarkers (NSP activity). However, the absolute magnitude of lung function benefit was limited and its clinical relevance remains uncertain; QoL effects were below the proposed MWPC threshold and were not alpha-controlled. Severe PE rates were numerically reduced but without statistical significance. Subgroup analyses indicated possible heterogeneity, with reduced efficacy in patients with FEV_1 <50%, baseline eosinophilia, asthma, COPD, or P. aeruginosa colonisation, and greater benefit in those with FEV_1 ≥50% and in Asian populations. A post hoc interaction for FEV_1 <50% suggested heterogeneity (p=0.0053 for 25 mg vs placebo), though numbers were small and analyses exploratory. These findings should be interpreted cautiously.

In the adolescent population, there is a limited small sample size and the lack of robust Bayesian operating characteristics. However, the extrapolation of efficacy to adolescents is supported by comparable pharmacokinetics similarility of the disease between both adult and adolescent populations.

The applicant has selected the 25 mg QD dose, based on broader and more consistent effects on lung function, biomarkers and exploratory QoL endpoints, despite comparable PE reductions at both doses. The CHMP considered this justification acceptable, while noting that the clinical relevance of the observed FEV_1 effect remains unclear. The 25 mg dose is therefore the agreed dos by CHMP.

Study INS1007-201 (WILLOW) data are supportive of the pivotal study. The patient population was similar to the one in pivotal study, and representative of the target population, but did not enroll adolescents. Efficacy endpoints were similar but measured over shorter period of time (24 vs 52 weeks in the pivotal study). PEs had a minimum of 4 weeks required between onset. The treatment effect was shown for both doses of brensocatib in comparison to placebo with a statistically significant result for the primary endpoint, the median time to first exacerbation.

In summary, the ASPEN study provides robust evidence of efficacy for brensocatib in reducing pulmonary exacerbations in patients with NCFBE and frequent prior PEs, with supportive findings from secondary endpoints and the WILLOW study.

In summary the CHMP agreed with the following indication: for the treatment of non-cystic fibrosis bronchiectasis (NCFB) in patients 12 years of age and older with two or more exacerbations in the prior 12 months.

6.4. Clinical safety

For the purpose of this document, the following definitions apply:

'Adverse event – AE' means any untoward medical occurrence in a subject to whom a medicinal product is administered and which does not necessarily have a causal relationship with this treatment.

'Serious adverse event – SAE' means any untoward medical occurrence that at any dose requires inpatient hospitalisation or prolongation of existing hospitalisation, results in persistent or significant disability or incapacity, results in a congenital anomaly or birth defect, is life-threatening, or results in death. The definition (in line with ICH E2A) includes important medical events that may not be immediately life-threatening or result in death or hospitalisation but may jeopardise the patient or may require intervention to prevent one of the other outcomes listed in the definition above.

'Adverse drug reaction – ADR' means any untoward and unintended response to a medicinal product related to any dose administered, for which, after a thorough assessment, a causal relationship between the medicinal product and the adverse event is at least a reasonable possibility, based for example, on their comparative incidence in clinical trials, or findings from epidemiological studies and/or on an evaluation of causality from individual case reports.

6.4.1. Safety data collection

The primary safety evaluation consists of pooled data from studies INS1007-201 and INS1007-301.

According to the Schedules of Assessments data was collected during safety visits at Day 1, weeks 2, 4, 8, 12, 16, 20, 24 (EOT) and 28 [INS1007-201] and at Day 1 (BL), weeks 4, 10 (phone), 16, 22 (phone), 28, 34 (phone), 40, 46 (phone), 52 (EOT), 56 (FU, EOS) [INS1007-301].

EOT=end of treatment, BL=baseline, FU=follow up, EOS=end of study

6.4.2. Patient exposure

Pooled studies INS1007-201 and INS1007-201

The primary safety evaluation consists of pooled data from 2 completed, randomized (1:1:1) placebo-controlled studies (placebo, 10 or 25 mg OD) in participants with NCFBE with at least 2 pulmonary exacerbations in the past 12 months:

INS1007-201 Phase 2 [WILLOW]: Adult patients (18 – 85 years) were treated for 24 weeks

INS1007-301 Phase 3 [ASPEN]: Adult patients (18 – 85 years) and adolescents (12-18 years, randomized 2:2:1) were treated for 52 weeks,

Table 77: Patient exposure - pooled studies

Study	Dose and Schedule	Exposure					Disposition
		Total Participants Exposed n	<12 weeks n	≥12 weeks n	≥24 weeks n	≥36 weeks n	52 Week Study Treatment Completed[a] n
Phase 2/3 - NCFBE							
Phase 3 INS1007-301	10 mg QD for 52 weeks	582	17	565	548	528	510
	25 mg QD for 52 weeks	574	17	557	538	527	512
	Total/adolescents	1156/33	34/1	1122/32	1086/32	1055/31	1022/31
Phase 2 INS1007-201	10 mg QD for 24 weeks	81	8	73	52	0	N/A
	25 mg QD for 24 weeks	89	10	79	57	0	N/A
	Total	170	18	152	109	0	N/A
Pooled INS1007-301 and INS1007-201 (Integrated Population for Safety Analysis)	10 mg	663	25	638	600	528	580
	25 mg	663	27	636	595	527	586
	Total Pooled	1326	52	1274	1195	1055	1166

Source: Module 2.7.4. Table 6

Table 78: Patient exposure in placebo controlled studies

	Patients enrolled	Patients exposed to any dose*	Patients exposed to the proposed dose (25mg)	Patients with long term** safety data (25mg)	Patients with long term** safety data (10mg)
Blinded studies (placebo-controlled)	1.974	1.326	663	595	600

* Received at least 1 dose of active treatment
** 6 months or 12 months continuous exposure data

Supportive studies (not pooled)

Safety data was derived from 10 completed Phase 1 studies in healthy participants (including participants with renal or hepatic impairment), a Phase 2 study in participants with CF, an ongoing Phase 2 study in participants with CRSsNP, and a supportive Phase 1 Study INS1007-110, which was completed after the data cutoff (28 March 2024).

Table 79: Exposure – supportive studies

Clinical study	Patient population	Dosing	Patients exposed to any dose* <12 weeks [n]
D6190C00001 SAD Part 1a/1b[a] MAD Part 2	Healthy adult subjects	5, 15, 35, 50, 65 mg 10, 25, 40 mg	30 24
D6190C00003	Healthy adult subjects	25 mg (SD)	15
INS1007-101	Healthy adult subjects (Caucasian, Japanese)	10, 25,40 mg	69
INS1007-102	Healthy adult subjects Subjects with renal impairment	25 mg (SD)	10 18
INS1007-103	Healthy adult male subjects	40 mg (SD)	7
INS1007-104	Healthy adult subjects	40, 80, 120 mg	44
INS1007-105	Healthy adult subjects Subjects with hepatic impairment	25 mg (SD)	9 18
INS1007-106	Healthy adult subjects	25 mg (SD on days 1 and 12 or 17)	32
INS1007-109	Healthy adult subjects	25 mg (SD on Days 1 and 13)	22
INS 1007-110[b]	Healthy adult subjects	25 mg (SD on Days 1 and 10)	24
INS1007-211	Patients with Cystic Fibrosis	10, 25, 40 mg	24
INS1007-221[c]	Adult patients with CRSsNP		15
Total			**346**

CRSsNP = chronic rhinosinusitis without nasal polyps; MAD = multiple ascending dose; NCFBE = non-cystic fibrosis bronchiectasis; SAD = single ascending dose.
a Eight subjects from Part 1a were rolled to Part 1b, of which five received brensocatib and three received placebo. Those subjects are counted once in the total of subjects.
b Although INS1007-110 started on 02 April 2024 (Final Clinical Study Report: 27 September 2024), i.e. after the primary analysis cutoff date of INS1007-301 (28 March 2024), it is included in Table 2 for completeness.
c Includes 15 of 23 subjects on blinded medication through 28 March 2024 since 2/3 subjects were exposed to brensocatib in this ongoing study
Scource: Module 1.8.2 Table 2

In addition, at data cutoff date 11-Jan-2025, 486 participants (12 adolescents) were enrolled in the open-label EAP (expanded access program) of Studies INS1007-301 (n = 458) and INS1007-201 (n= 28). The average length of exposure within the EAP was 954 days for the 23 participants on brensocatib 25 mg/day and 432 days for the 463 participants on brensocatib 10 mg/day.

6.4.3. Adverse events

Table 80: Studies INS1007-301 and INS1007-201: Overall Summary of Treatment-Emergent Adverse Events (Safety Analysis Set)

Characteristics	Brensocatib 10 mg N=663 N (%)	Brensocatib 25 mg N=663 N (%)	Brensocatib pooled N=1326 N (%)	Placebo N=648 N (%)
Any TEAE	528 (79.6)	516 (77.8)	1044 (78.7)	515 (79.5)
Related TEAE	110 (16.6)	122 (18.4)	232 (17.5)	108 (16.7)
Serious TEAE	113 (17.0)	107 (16.1)	220 (16.6)	127 (19.6)
Serious related TEAE	3 (0.5)	1 (0.2)	4 (0.3)	5 (0.8)
TEAE resulting in Death	3 (0.5)	5 (0.8)	8 (0.6)	7 (1.1)
TEAE leading to study treatment withdrawal	31 (4.7)	28 (4.2)	59 (4.4)	32 (4.9)
TEAE leading to study withdrawal	17 (2.6)	20 (3.0)	37 (2.8)	19 (2.9)

Scource: Module 2.7.4. Table 11

Table 81: Studies INS1007-301 and INS1007-201: Summary of Treatment-Emergent Adverse Events in >=1% Participants in Any Treatment Group, System Organ Class and Preferred Term (Safety Analysis Set)

System Organ Class Preferred Term	**Brensocatib 10 mg QD (N=663)**	**Brensocatib 25 mg QD (N=663)**	**Pooled Brensocatib (N=1326)**	**Placebo (N=648)**
	n (%)	**n (%)**	**n (%)**	**n (%)**
Participants with at least one qualified event	528 (79.6)	516 (77.8)	1044 (78.7)	515 (79.5)
Infections and infestations	**338 (51.0)**	**335 (50.5)**	**673 (50.8)**	**337 (52.0)**
COVID-19	92 (13.9)	120 (18.1)	212 (16.0)	89 (13.7)
Nasopharyngitis	49 (7.4)	38 (5.7)	87 (6.6)	46 (7.1)
Urinary tract infection	30 (4.5)	35 (5.3)	65 (4.9)	37 (5.7)
Sinusitis	34 (5.1)	29 (4.4)	63 (4.8)	34 (5.2)
Upper respiratory tract infection	30 (4.5)	26 (3.9)	56 (4.2)	20 (3.1)
Pneumonia	26 (3.9)	27 (4.1)	53 (4.0)	31 (4.8)
Influenza	27 (4.1)	10 (1.5)	37 (2.8)	26 (4.0)
Bronchitis	9 (1.4)	9 (1.4)	18 (1.4)	15 (2.3)
Gastroenteritis	10 (1.5)	8 (1.2)	18 (1.4)	3 (0.5)
Oral candidiasis	11 (1.7)	7 (1.1)	18 (1.4)	6 (0.9)
Rhinitis	10 (1.5)	7 (1.1)	17 (1.3)	7 (1.1)
Viral infection	8 (1.2)	8 (1.2)	16 (1.2)	8 (1.2)
Acute sinusitis	9 (1.4)	6 (0.9)	15 (1.1)	4 (0.6)
Herpes zoster	4 (0.6)	11 (1.7)	15 (1.1)	7 (1.1)
Conjunctivitis	6 (0.9)	8 (1.2)	14 (1.1)	4 (0.6)
Pharyngitis	7 (1.1)	6 (0.9)	13 (1.0)	5 (0.8)
Ear infection	8 (1.2)	3 (0.5)	11 (0.8)	6 (0.9)
Infective exacerbation of bronchiectasis	5 (0.8)	4 (0.6)	9 (0.7)	9 (1.4)
Pseudomonas infection	3 (0.5)	5 (0.8)	8 (0.6)	9 (1.4)
Respiratory, thoracic and mediastinal disorders	**196 (29.6)**	**189 (28.5)**	**385 (29.0)**	**203 (31.3)**

System Organ Class Preferred Term	Brensocatib 10 mg QD (N=663)	Brensocatib 25 mg QD (N=663)	Pooled Brensocatib (N=1326)	Placebo (N=648)
	n (%)	n (%)	n (%)	n (%)
Cough	56 (8.4)	47 (7.1)	103 (7.8)	46 (7.1)
Bronchiectasis	50 (7.5)	49 (7.4)	99 (7.5)	67 (10.3)
Haemoptysis	25 (3.8)	19 (2.9)	44 (3.3)	28 (4.3)
Dyspnoea	20 (3.0)	24 (3.6)	44 (3.3)	24 (3.7)
Sputum increased	19 (2.9)	20 (3.0)	39 (2.9)	16 (2.5)
Oropharyngeal pain	14 (2.1)	16 (2.4)	30 (2.3)	11 (1.7)
Rhinitis allergic	8 (1.2)	8 (1.2)	16 (1.2)	6 (0.9)
Wheezing	6 (0.9)	7 (1.1)	13 (1.0)	6 (0.9)
Epistaxis	4 (0.6)	8 (1.2)	12 (0.9)	4 (0.6)
Rhinorrhoea	5 (0.8)	7 (1.1)	12 (0.9)	17 (2.6)
Productive cough	7 (1.1)	2 (0.3)	9 (0.7)	10 (1.5)
Gastrointestinal disorders	**168 (25.3)**	**148 (22.3)**	**316 (23.8)**	**176 (27.2)**
Diarrhoea	31 (4.7)	24 (3.6)	55 (4.1)	36 (5.6)
Nausea	15 (2.3)	13 (2.0)	28 (2.1)	25 (3.9)
Constipation	15 (2.3)	11 (1.7)	26 (2.0)	15 (2.3)
Abdominal pain	11 (1.7)	8 (1.2)	19 (1.4)	10 (1.5)
Gastrooesophageal reflux disease	7 (1.1)	11 (1.7)	18 (1.4)	9 (1.4)
Gingival disorder	10 (1.5)	8 (1.2)	18 (1.4)	9 (1.4)
Gingival pain	8 (1.2)	9 (1.4)	17 (1.3)	13 (2.0)
Vomiting	7 (1.1)	9 (1.4)	16 (1.2)	14 (2.2)
Toothache	5 (0.8)	10 (1.5)	15 (1.1)	7 (1.1)
Abdominal pain upper	9 (1.4)	3 (0.5)	12 (0.9)	9 (1.4)
Dyspepsia	9 (1.4)	3 (0.5)	12 (0.9)	9 (1.4)
Gingival bleeding	9 (1.4)	3 (0.5)	12 (0.9)	4 (0.6)
Dry mouth	5 (0.8)	5 (0.8)	10 (0.8)	9 (1.4)
Dental caries	5 (0.8)	2 (0.3)	7 (0.5)	9 (1.4)
Skin and subcutaneous tissue disorders	**113 (17.0)**	**143 (21.6)**	**256 (19.3)**	**106 (16.4)**
Rash	19 (2.9)	27 (4.1)	46 (3.5)	15 (2.3)
Dry skin	12 (1.8)	20 (3.0)	32 (2.4)	11 (1.7)
Hyperkeratosis	8 (1.2)	16 (2.4)	24 (1.8)	4 (0.6)
Eczema	11 (1.7)	11 (1.7)	22 (1.7)	8 (1.2)
Pruritus	11 (1.7)	11 (1.7)	22 (1.7)	13 (2.0)
Alopecia	7 (1.1)	8 (1.2)	15 (1.1)	3 (0.5)
Skin exfoliation	8 (1.2)	7 (1.1)	15 (1.1)	0
Dermatitis	6 (0.9)	8 (1.2)	14 (1.1)	1 (0.2)
Erythema	8 (1.2)	5 (0.8)	13 (1.0)	4 (0.6)
Skin lesion	4 (0.6)	9 (1.4)	13 (1.0)	6 (0.9)
Urticaria	1 (0.2)	12 (1.8)	13 (1.0)	3 (0.5)
Musculoskeletal and connective tissue disorders	**106 (16.0)**	**95 (14.3)**	**201 (15.2)**	**127 (19.6)**
Arthralgia	31 (4.7)	19 (2.9)	50 (3.8)	24 (3.7)
Back pain	14 (2.1)	20 (3.0)	34 (2.6)	38 (5.9)
Pain in extremity	15 (2.3)	9 (1.4)	24 (1.8)	20 (3.1)
Myalgia	6 (0.9)	9 (1.4)	15 (1.1)	9 (1.4)
Muscle spasms	6 (0.9)	7 (1.1)	13 (1.0)	6 (0.9)
Osteoarthritis	4 (0.6)	8 (1.2)	12 (0.9)	7 (1.1)

System Organ Class Preferred Term	Brensocatib 10 mg QD (N=663)	Brensocatib 25 mg QD (N=663)	Pooled Brensocatib (N=1326)	Placebo (N=648)
	n (%)	n (%)	n (%)	n (%)
Musculoskeletal chest pain	1 (0.2)	3 (0.5)	4 (0.3)	7 (1.1)
General disorders and administration site conditions	**95 (14.3)**	**102 (15.4)**	**197 (14.9)**	**99 (15.3)**
Pyrexia	28 (4.2)	30 (4.5)	58 (4.4)	27 (4.2)
Fatigue	22 (3.3)	29 (4.4)	51 (3.8)	27 (4.2)
Non-cardiac chest pain	12 (1.8)	10 (1.5)	22 (1.7)	13 (2.0)
Asthenia	8 (1.2)	8 (1.2)	16 (1.2)	8 (1.2)
Malaise	9 (1.4)	7 (1.1)	16 (1.2)	4 (0.6)
Chest discomfort	6 (0.9)	7 (1.1)	13 (1.0)	2 (0.3)
Oedema peripheral	5 (0.8)	7 (1.1)	12 (0.9)	4 (0.6)
Pain	8 (1.2)	3 (0.5)	11 (0.8)	6 (0.9)
Chest pain	2 (0.3)	4 (0.6)	6 (0.5)	9 (1.4)
Nervous system disorders	**91 (13.7)**	**104 (15.7)**	**195 (14.7)**	**84 (13.0)**
Headache	47 (7.1)	61 (9.2)	108 (8.1)	42 (6.5)
Dizziness	14 (2.1)	16 (2.4)	30 (2.3)	12 (1.9)
Migraine	7 (1.1)	3 (0.5)	10 (0.8)	2 (0.3)
Injury, poisoning and procedural complications	**71 (10.7)**	**73 (11.0)**	**144 (10.9)**	**73 (11.3)**
Tooth fracture	7 (1.1)	8 (1.2)	15 (1.1)	8 (1.2)
Fall	9 (1.4)	4 (0.6)	13 (1.0)	11 (1.7)
Contusion	7 (1.1)	5 (0.8)	12 (0.9)	10 (1.5)
Investigations	**48 (7.2)**	**49 (7.4)**	**97 (7.3)**	**62 (9.6)**
Weight decreased	4 (0.6)	8 (1.2)	12 (0.9)	11 (1.7)
Vascular disorders	**50 (7.5)**	**25 (3.8)**	**75 (5.7)**	**34 (5.2)**
Hypertension	30 (4.5)	14 (2.1)	44 (3.3)	22 (3.4)
Metabolism and nutrition disorders	**40 (6.0)**	**34 (5.1)**	**74 (5.6)**	**40 (6.2)**
Hyperglycaemia	7 (1.1)	0	7 (0.5)	3 (0.5)
Cardiac disorders	**25 (3.8)**	**30 (4.5)**	**55 (4.1)**	**23 (3.5)**
Atrial fibrillation	5 (0.8)	7 (1.1)	12 (0.9)	8 (1.2)
Ear and labyrinth disorders	**17 (2.6)**	**24 (3.6)**	**41 (3.1)**	**28 (4.3)**
Vertigo	5 (0.8)	6 (0.9)	11 (0.8)	12 (1.9)
Psychiatric disorders	**22 (3.3)**	**18 (2.7)**	**40 (3.0)**	**23 (3.5)**
Insomnia	5 (0.8)	7 (1.1)	12 (0.9)	9 (1.4)
Immune system disorders	**17 (2.6)**	**12 (1.8)**	**29 (2.2)**	**11 (1.7)**
Seasonal allergy	7 (1.1)	5 (0.8)	12 (0.9)	2 (0.3)

Scource: Module 2.7.4. Table 12

Table 82: Studies INS1007-301 and INS1007-201: Summary of Severe Treatment-Emergent Adverse Events in ≥2% Participants in Any Treatment Group

System Organ Class Preferred Term	Severity	Brensocatib 10 mg QD (N = 663) n (%)	Brensocatib 25 mg QD (N = 663) n (%)	Pooled Brensocatib (N = 1326) n (%)	Placebo (N = 648) n (%)
Participants with at least one qualified event	Mild	438 (66.1)	422 (63.7)	860 (64.9)	419 (64.7)
	Moderate	305 (46.0)	285 (43.0)	590 (44.5)	275 (42.4)
	Severe	79 (11.9)	74 (11.2)	153 (11.5)	103 (15.9)

Scource: Module 2.7.4. Table 15

The proportion of participants reporting any TEAEs was similar across all treatment groups (10 mg, 25 mg, and placebo). The majority of TEAEs were of mild or moderate intensity.

Most TEAEs belong to the SOC infections and infestations with COVID-19 being the most frequent TEAE in all treatment groups. The incidence was higher in the brensocatib groups, also upper respiratory tract infections (URTI) and gastroenteritis were reported at a higher frequency (>1% difference) in any of the brensocatib groups. Several similar PTs (e.g. viral URTI, pharyngitis or gastroenteritis viral, gastroenteritis baceterial) were reported separately (incidences partly < 1% in any treatment group). Despite the immune-modulatory MoA of brensocatib no further TEAE frequencies of this SOC separated clearly from placebo. However, when pooling opportunistic infections, these seem to occur with a modest increase in brensocatib groups compared to placebo with no clear dose-dependency. The same accounts for candida infections.

Both brensocatib doses were associated with a higher frequency of TEAEs belonging to the **SOC** Skin and subcutaneous tissue disorders with the 25 mg dose generally showing the highest rates. The most common events were rash, dry skin, hyperkeratosis, pruritus and skin exfoliation. Dermatitis events occurred at lower frequencies, but more frequent (> 1% difference) compared to placebo. The rates of alopecia (PTs alopecia and diffuse alopecia) were higher in both brensocatib groups compared to placebo. Events of hair texture abnormal and hair growth abnormal were reported only in brensocatib groups, albeit with low frequencies. Additionally, PTs referring to pigmentation disorders (pigmentation disorder, skin hyperpigmentation, nail discolouration, skin discolouration, lentigo) were reported almost exclusively with brensocatib (11 events in brensocatib groups vs 1 in placebo).

Overall, in the SOC Neoplasms benign, malignant and unspecified (including cysts and polyps) TEAEs were reported in 3.0%, 3.9%, and 2.3% of participants for brensocatib 10 mg, 25 mg, and placebo groups respectively. No TEAEs in this SOC showed a difference in incidence ≥1.0% between either brensocatib and placebo group.

The overall incidence of TEAEs of the SOC Nervous system disorders was higher in the brensocatib 25 mg group compared to placebo, with headache as the most frequently reported individual adverse event across all treatment groups and demonstrating dose ordering, affecting 7.1% of patients in the brensocatib 10 mg group, 9.2% in the 25 mg group, and 6.5% in the placebo group. A similar trend was noted also for dizziness and migraine. Most other adverse events occurred at low frequencies, typically affecting fewer than 1% of subjects in each treatment group, with many events showing isolated occurrences in single subjects across the study population. Five events of syncope and 2 events of loss of consciousness occurred in brensocatib group, with 0 events in placebo.

Seasonal allergy (SOC Immune system disorders) was also reported more frequent compared to placebo.

6.4.3.1. Adverse drug reactions

The primary assessment for ADRs was conducted on the pooled safety analysis set of Studies INS1007-301 and INS1007-201. For ADR identification, treatment-emergent adverse events which were more frequent on both brensocatib groups than placebo (preferred terms ≥2%) and more frequent in the brensocatib 25 group than placebo (≥1% higher incidence) were further analyzed to determine the clinical relevance and for plausibility of a causal association with treatment with brensocatib. In addition, other TEAEs were also assessed by dose-response trends, severity, causality, seriousness, leading to death and treatment discontinuation to contextualize ADR identification. The agreed ADRs are described later in section 6.4.11.1.2.

6.4.4. Adverse events of special interest, serious adverse events and deaths, other significant events

Adverse events of special interest:

AESIs were prospectively defined and monitored during the conduct of clinical studies INS1007-301 and INS1007-201. For the pooled dataset clustered terms (SMQ or HLT) and individual preferred terms considered as Sponsor customized MedDRA query (CMQ) terms were used.

Table 83: Studies INS1007-301 and INS1007-201: Statistical Analysis of Treatment-Emergent Adverse Events of Special Interest by Investigator and by CMQ (Safety Analysis Set)

Characteristics	Brensocatib 10 mg QD (N = 663) n (%)	Brensocatib 25 mg QD (N = 663) n (%)	Placebo (N = 648) n (%)
TEAE of Special Interest by Investigator			
Any TEAE of special interest by investigator	59 (8.9)	79 (11.9)	66 (10.2)
Hyperkeratosis	20 (3.0)	38 (5.7)	14 (2.2)
Periodontal/gingival	21 (3.2)	21 (3.2)	18 (2.8)
Pneumonia	24 (3.6)	32 (4.8)	37 (5.7)
TEAE of Special Interest by CMQ			
Any TEAE of special interest by CMQ	64 (9.7)	65 (9.8)	56 (8.6)
Hyperkeratosis	29 (4.4)	39 (5.9)	20 (3.1)
Periodontal/gingival	39 (5.9)	35 (5.3)	38 (5.9)

The individual preferred terms (PT) considered as Sponsor customized MedDRA query (CMQ) term hyperkeratosis included all PTs of skin lesion, exfoliative rash, seborrheic keratosis, eczema, palmoplantar keratoderma and hyperkeratosis follicularis et parafollicularis.

The overall incidence of AESIs reported in each category (reported by Investigator and by CMQ) was low across treatment groups.

The most frequently reported preferred terms per CMQ (periodontal/gingival) were gingival disorder (10 [1.5%], 8 [1.2%], and 9 [1.4%]); periodontal disease was reported at lower frequencies (2 [0.3%], 5 [0.8%], and 1 [0.2%], for brensocatib 10 mg, brensocatib 25 mg, and placebo groups, respectively).

Serious Adverse Events:

Table 84: Studies INS1007-301 and INS1007-201: Summary of Serious Treatment-Emergent Adverse Events in ≥2 Participants in Any Brensocatib Group and More Frequent than Placebo by System Organ Class and Preferred Term (Safety Analysis Set)

System Organ Class Preferred Term	Brensocatib 10 mg QD (N = 663) n (%)	Brensocatib 25 mg QD (N = 663) n (%)	Pooled Brensocatib (N = 1326) n (%)	Placebo (N = 648) n (%)
Participants with at least one TEAE	**113 (17.0)**	**107 (16.1)**	**220 (16.6)**	**127 (19.6)**
Infections and infestations	**43 (6.5)**	**46 (6.9)**	**89 (6.7)**	**47 (7.3)**
COVID-19	4 (0.6)	9 (1.4)	13 (1.0)	6 (0.9)
Urinary tract infection	1 (0.2)	2 (0.3)	3 (0.2)	1 (0.2)
Appendicitis	2 (0.3)	0	2 (0.2)	1 (0.2)
Herpes zoster	0	2 (0.3)	2 (0.2)	1 (0.2)
Lung abscess	0	2 (0.3)	2 (0.2)	1 (0.2)
Sinusitis	2 (0.3)	0	2 (0.2)	0
Injury, poisoning and procedural complications	**11 (1.7)**	**4 (0.6)**	**15 (1.1)**	**7 (1.1)**
Joint dislocation	2 (0.3)	0	2 (0.2)	0
Meniscus injury	2 (0.3)	0	2 (0.2)	0
Gastrointestinal disorders	2 (0.3)	6 (0.9)	8 (0.6)	8 (1.2)
Intestinal obstruction	1 (0.2)	2 (0.3)	3 (0.2)	0

Scource: Module 2.7.4 Table 17

The overall incidence of SAEs was similar across treatment groups. The most frequently reported SAEs were bronchiectasis and pneumonia, and both were more frequent in the placebo group.

The only SAE reported in >2 participants in any brensocatib group and more frequent than placebo was COVID-19. All other SAEs more frequent in the brensocatib groups than placebo were reported in 1 or 2 participants each.

Frequency of SAEs deemed treatment-related was low.

Table 85: Studies INS1007-301 and INS1007-201: Summary of Serious Treatment-Emergent Adverse Events judged as related to treatment

brensocatib 10 mg n=3 [0.5%]	brensocatib 25 mg n=1 [0.2%]	Placebo N=5 [0.8%]
lymphocytosis	eosinophilic pneumonia	colitis
infective exacerbation of bronchiectasis		biliary colic
transient ischaemic attack		infective exacerbation of bronchiectasis
		pneumonia bacterial
		lung infiltration
		respiratory failure

Deaths:

Table 86: Studies INS1007-301 and INS1007-201: Summary of Treatment-Emergent Adverse Events Resulting in Death by System Organ Class and Preferred Term (Safety Analysis Set)

System Organ Class Preferred Term	Brensocatib 10 mg QD (N = 663) n (%)	Brensocatib 25 mg QD (N = 663) n (%)	Pooled Brensocatib (N = 1326) n (%)	Placebo (N = 648) n (%)
Participants with at least one TEAE, n (%)	3 (0.5)	5 (0.8)	8 (0.6)	7 (1.1)
Respiratory, thoracic and mediastinal disorders	**2 (0.3)**	**1 (0.2)**	**3 (0.2)**	**3 (0.5)**
Acute respiratory failure	1 (0.2)	0	1 (0.1)	1 (0.2)
Bronchiectasis	1 (0.2)	0	1 (0.1)	1 (0.2)
Respiratory failure	0	1 (0.2)	1 (0.1)	0
Haemoptysis	0	0	0	1 (0.2)
Infections and infestations	**1 (0.2)**	**1 (0.2)**	**2 (0.2)**	**1 (0.2)**
Aspergillus infection	1 (0.2)	0	1 (0.1)	0
Pneumonia	0	1 (0.2)	1 (0.1)	1 (0.2)
Cardiac disorders	0	1 (0.2)	1 (0.1)	2 (0.3)
Myocardial infarction	0	1 (0.2)	1 (0.1)	0
Cardiac arrest	0	0	0	1 (0.2)
Cardio-respiratory arrest	0	0	0	1 (0.2)
General disorders and administration site conditions	**0**	**1 (0.2)**	**1 (0.1)**	**0**
General physical health deterioration	0	1 (0.2)	1 (0.1)	0
Injury, poisoning and procedural complications	0	1 (0.2)	1 (0.1)	1 (0.2)
Road traffic accident	0	1 (0.2)	1 (0.1)	0
Cervical vertebral fracture	0	0	0	1 (0.2)

Scource: Module 2.7.4. Table 16

Fifteen participants had a fatal TEAE during the treatment period. Additionally, 1 participant had a post-treatment fatal AE of respiratory failure in Study INS1007-301 (placebo group). 4 deaths were reported in the EAP.

The rate of deaths was lower in participants treated with brensocatib in comparison to placebo.

None of the fatal AEs were deemed related to study treatment.

6.4.5. Discontinuation due to adverse events

The most common TEAE leading to study discontinuation was pneumonia (1 [0.2%], 1 [0.2%], and 3 [0.5%] for brensocatib 10 mg, brensocatib 25 mg, and placebo, respectively), followed by headache (1 [0.2%], 2 [0.3%], and 0), diarrhoea (2 [0.3%], 0, 0), nausea (0, 2 [0.3%], 0) and fatigue (0, 2 [0.3%], 0).

Table 87: Studies INS1007-301 and INS1007-201: Treatment Emergent Adverse Events Leading to Discontinuation

	Brensocatib 10 mg N=663 n (%)	Brensocatib 25 mg N=663 n (%)	Pooled Brensocatib N=1326 n %)	Placebo N=648 n (%)
TEAEs leading to treatment discontinuation	31 (4.7)	28 (4.2)	59 (4.4)	32 (4.9)
TEAEs leading to study discontinuation	17 (2.6)	20 (3.0)	37 (2.8)	19 (2.9)
TEAEs leading to dose interruptions	52 (7.8)	70 (10.6)	122 (9.2)	63 (9.7)
Related TEAEs leading to treatment discontinuation	31 (4.7)	28 (4.2)	59 (4.4)	32 (4.9)
Related TEAEs leading to study discontinuation	17 (2.6)	20 (3.0)	37 (2.8)	19 (2.9)
SAEs leading to treatment discontinuation	9 (1.4)	7 (1.1)	16(1.2)	14 (2.2)
SAEs leading to study discontinuation	7 (1.1)	7 (1.1)	14 (1.1)	10 (1.5)

Overall, frequency of TEAEs leading to treatment discontinuation was low and similar between groups (<5% in any treatment group). The most common TEAEs leading to discontinuation were bronchiectasis and headache. No meaningful differences between treatment groups were observed for any TEAE leading to treatment discontinuation, except for the ADR headache (2 [0.3%], 5 [0.8%], and 0 for brensocatib 10 mg, brensocatib 25 mg, and placebo, respectively). ADRs belonging to the SOC skin and subcutaneous tissue disorders overall led to treatment discontinuation only in the brensocatib groups (pooled 7 [0.7%] vs placebo: 0).

Four additional events of treatment discontinuation were reported in the supportive studies due to Lichen planus like drug eruption, dyspnea and dry throat, anaemia and severe HI (Child-Pugh 10-15 group), hyponatremia.

6.4.6. Safety in special populations

AEs by age range

Table 88: Summary of TEAEs and SAEs of Participants by Age (Safety Analysis Set)

Preferred Term	Age (years)	Brensocatib 10 mg QD n (%)	Brensocatib 25 mg QD n (%)	Pooled Brensocatib n (%)	Placebo n (%)
Number of participants	12 to <18	17	16	33	8
	≥18	646	647	1293	640
	18 to <65	322	295	617	328
	≥65	324	352	676	312
	<75	560	565	1125	542
	≥75	103	98	201	106
Participants with at least one qualified event of TEAE	12 to <18	15 (88.2)	12 (75.0)	27 (81.8)	7 (87.5)
	≥18	513 (79.4)	504 (77.9)	1017 (78.7)	508 (79.4)
	18 to <65	259 (80.4)	223 (75.6)	482 (78.1)	258 (78.7)
	≥65	254 (78.4)	281 (79.8)	535 (79.1)	250 (80.1)
	<75	444 (79.3)	438 (77.5)	882 (78.4)	427 (78.8)
	≥75	84 (81.6)	78 (79.6)	162 (80.6)	88 (83.0)
Participants with at least one qualified event of SAE	12 to <18	2 (11.8)	3 (18.8)	5 (15.2	1 (12.5)
	>18	111 (17.2	104 (16.1)	215 (16.6)	126 (19.7)
	18 to <65	50 (15.5)	44 (14.9)	94 (15.2)	53 (6.2)
	≥65	61 (18.8)	60 (17.0)	121 (17.9)	73 (23.4)
	<75	95 (17.0)	86 (15.2)	181 (16.1)	96 (17.7)
	≥75	18 (17.5)	21 (21.4)	39 (19.4)	31 (29.2)

Scource: Module 2.7.4. Table 34 and 35, Updated ISS Tables 1.8-2a and 1.8-2b.

Adult (≥18 and ≤85 years) and adolescent (≥12 and <18 years) participants were included in studies INS1007-201 and INS1007-301.

After INS1007-301 study completion a total of 15 (88.2%), 12 (75.0%), and 7 (87.5%) adolescents in the brensocatib 10 mg, brensocatib 25 mg, and placebo groups, respectively, had at least 1 TEAE. The most common TEAEs reported for ≥2 adolescent participants in the brensocatib 10 or 25 mg groups that were more frequent than placebo were similar to the primary analysis and included URTI (3 [17.6%], 2 [12.5%], and 0 in the brensocatib 10 mg, brensocatib 25 mg, and placebo groups, respectively), gingival disorder (1 [5.9%], 2 [12.5%], and 0), diarrhea (2 [11.8%], 0, and 0), gastroenteritis (0, 2 [12.5%], and 0), viral infection (0, 2 [12.5%], and 0), viral pharyngitis (2 [11.8%], 0, and 0), and skin lesion (0, 2 [12.5%], and 0).

As regards the elderly population (≥65 years), the frequency of TEAEs is comparable to the adult population (< 65 years), however, SAEs were observed at a slightly higher frequency compared to adults < 65 years.

No clinically meaningful differences in TEAE or SAE frequency reported for sex, BMI, race, BSI score and region.

In the ethnic subgroup of Hispanic or Latino (26.1% of participants) reported TEAEs, and SAEs were approximately 20% lower in all treatment groups, maintaining the overall pattern of TEAE.

Higher frequency of TEAEs were observed in all treatment groups for participants with chronic use of inhaled antibiotics, chronic use of macrolides, and smoking. Incidence of pneumonia and SAEs was higher in participants with baseline ICS versus participants without across all treatment groups, more pronounced in the placebo groups.

6.4.7. Immunological events

Not applicable

6.4.8. Safety related to drug-drug interactions and other interactions

No potential impact of CYP3A4, Pgp or other interactions on safety has been observed in studies INS1007-301 and INS1007-201, in which concomitant medications were permitted per protocols.

6.4.9. Vital signs and laboratory findings

Hematology, clinical chemistry, and urinalysis

No clinically meaningful trends or changes in clinical laboratory test values were observed with either dose of brensocatib. Hematology, clinical chemistry, and urinalysis findings showed no treatment group differences, with no dose-related trends. Frequency of clinical laboratory findings reported as TEAEs was low. The majority were of mild or moderate severity, not related to study treatment, nonserious, and resolved.

Metabolite Toxicity – Thiocyanate

The endogenous compound Thiocyanate was identified as a major metabolite. Brensocatib at 25 mg QD or up to 120 mg SD did not have any impact on thiocyanate exposure in healthy, renally or hepatically impaired, or NCFBE participants, the observed thiocyanate plasma levels were within the normal range. In Study INS1007-301, events of hypothyroidism, hypotension, and neurotoxicity were evaluated for their potential association with thiocyanate. No correlation was observed between thiocyanate concentrations and possible metabolite toxicity events.

Vital Signs

There were no clinically relevant changes in vital signs during the study, and no imbalance was observed for clinically meaningful changes from baseline across the treatment groups.

Electrocardiography

Incidence of ECG-related TEAEs was low and similar across treatment groups. In pooled studies, a QTcF interval >450 ms or a >30 ms change from baseline at any time during the study were reported for <10% of participants in any treatment group.

6.4.10. Post-marketing experience

Not applicable.

6.4.11. Overall discussion and conclusions on clinical safety

6.4.11.1. Discussion

Patient exposure

Overall, a total of 1.672 subjects has been exposed to brensocatib. The primary safety evaluation of this NAS consists of pooled data from 1.326 patients treated with 10 or 25 mg brensocatib in studies

INS1007-201 and -301 and is considered of sufficient size. 663 participants, including 16 adolescents, were treated with the proposed dose of brensocatib (25 mg). The safety database is considered large enough and the exposure duration (cut of 11 Jan 2025) is considered long enough to enable comprehensive assessment. In line with ICH E1, data on 52-week treatment are sufficient to support the intended chronic treatment. An open-label EAP (expanded access program) is ongoing and will provide further safety data beyond one year. As patients from the -301 study, however, proceed with the 10 mg dose the value of this long-term data is limited for the 25 mg to be marketed formulation.

Adverse events

Adverse event analysis from the pooled Studies INS1007-301 and INS1007-201 showed that participants experienced TEAEs with similar rates across the treatment groups. The majority of TEAEs were mild to moderate in severity. Frequency of severe TEAEs was generally low with both brensocatib groups reporting fewer severe TEAEs than the placebo group.

Most TEAEs belong to the SOC infections and infestations with COVID-19 being the most frequent TEAE in all treatment groups. Notably, the incidence of COVID-19 infections was higher in the brensocatib groups, also upper respiratory tract infections (URTI) and gastroenteritis were reported at higher frequencies in any of the brensocatib groups (>1% difference to placebo). Several similar PTs (e.g. viral URTI, pharyngitis or gastroenteritis viral, gastroenteritis baceterialbacterial) were reported seperately. Respective pooled analyses were provided revealing a group difference to placebo of > 1% for URTI as well as for gastroenteritis events. Regarding gastroenteritis, DPP1 inhibition may compromise gastrointestinal host defense, particularly against bacterial gastroenteritis, due to its impact on neutrophil serine protease activity, which is important for antimicrobial action and immune cell recruitment. This could result in impaired bacterial clearance and diminished local inflammatory responses as neutrophils play a central role in bacterial defense, while in viral and parasitic infections, other immune cells such as eosinophils may predominate. Therefore, this is included as missing information (Long-term safety (in particular the risk for infections and malignancies) in the RMP

No further TEAE frequencies of this SOC separated clearly from placebo. However, when pooling opportunistic infections, these seem to occur with a modest increase in brensocatib groups compared to placebo with no clear dose-dependency. The same accounts for candida infections. Although it is acknowledged that DPP1 inhibition with brensocatib does not cause generalized immune dysfunction, it selectively impairs neutrophil serine protease-dependent host defense leading to functional neutrophil defects despite normal neutrophil counts. This selective impairment may increase susceptibility to certain infections, especially those where neutrophil serine proteases are critical for host defense, such as invasive and fungal infections. The observed imbalance in fungal infections indicative of immunosuppression (oral candidiasis, oesophageal candidiasis, oropharyngeal candidiasis, Aspergillus infections, oral fungal infections, and fungal bronchitis) is addressed in Section 4.4 SmPC.

Both brensocatib doses were associated with a higher frequency of various skin and subcutaneous tissue adverse events with the 25 mg dose generally showing the highest rates. The most common events were hyperkeratosis, dermatitis, rash and dry skin, but alsoskin exfoliation and alopecia were identified as adverse events.

Adverse drug reactions

In summary, Headache (SOC nervous system disorder), gingival disorder, periodontal disorder (SOC gastrointestinal disorder) rash, dry skin, dermatitis, skin exfoliation, hyperkeratosis and alopecia (SOC skin and subcutanesous disorder), upper respiratory tract infection (URTI) and gastroenteritis (both SOC infections and infestations) have been identified as ADR by the applicant, which is supported.

Headache: There was a dose-ordering effect (47 [7.1%], 61 [9.2 %], and 42 [6.5%] for bresocatib 10 mg, brensocatib 25 mg, and placebo groups, respectively). Most headache events were mild or moderate, none were serious, and most participants recovered while on treatment. There were nine severe cases of headache. No action was taken with study treatment in these participants with severe events, and most of the participants recovered from the events.

Hyperkeratosis: The rate of hyperkeratosis was higher in each brensocatib group than placebo and showed dose ordering (8 [1.2%], 16 [2.4%], and 4 [0.6%] for brensocatib 10 mg, brensocatib 25 mg, and placebo groups, respectively). All events were nonserious, mild or moderate in severity, and most resolved with no action taken with study treatment. One event of hyperkeratosis of mild intensity in the brensocatib 25 mg group led to treatment discontinuation. Hyperkeratosis was included as ADR per AESI CMQ definition including also skin lesion, keratosis pilaris, exfoliative rash and seborrhoeic keratosis, the observed frequencies were respectively higher (29 [4.4%], 39 [5.9%], and 20 [3.1%] for brensocatib 10 mg, brensocatib 25 mg, and placebo groups, respectively).

Rash: The rate of rash was higher in each brensocatib group than placebo and showed dose ordering (19 [2.9%], 27 [4.1%], and 15 [2.3%] for brensocatib 10 mg, brensocatib 25 mg, and placebo groups, respectively). Most events were mild and not related, no event led to drug or study discontinuation, and all events were recovered by end of study.

Dry skin: While the 10 mg dose had the same frequency as that of the placebo group, the 25 mg group had a higher frequency to that of placebo (12 [1.8%], 20 [3.0%], and 11 [1.7%] of participants in the brensocatib 10 mg, brensocatib 25 mg, and placebo groups, respectively). All events were mild to moderate in severity, and no event led to study treatment interruption, study treatment discontinuation, or study discontinuation. Most cases were recovered by the end of the study.

Dermatitis: A pooled analysis of dermatitis events of multifactorial origin as well as eczema PTs shows an imbalance with the rate being higher in the active treatment groups (brensocatib 10 mg: 20 [3.0%], brensocatib 25 mg: 28 [4.2%] vs. placebo 15 [2.3%),

Alopecia: The rates of alopecia (PTs alopecia and diffuse alopecia) were higher (> 1%) in both brensocatib groups compared to placebo. Events of hair texture abnormal and hair growth abnormal were reported only in brensocatib groups, albeit with low frequencies.

Upper respiratory tract infections (URTI): Updated rates for URTI based on the complete safety data from INS1007-301 reveal a group difference to placebo of > 1% (30 (4.5%), 26 (3.9%), 56 (4.2%), 20 (3.1%) in the brensocatib 10 mg, brensocatib 25 mg, pooled (10 and 25 mg) and placebo groups.

Gastroenteritis: In addition to 2 severe events, the rate of gastroenteritis was more than two-fold higher in each brensocatib group than placebo (10 [1.5%], 8 [1.2%], and 3 [0.5%] for brensocatib 10 mg, brensocatib 25 mg, and placebo groups, respectively).

Based on the drug's mechanism of action (dipeptidyl peptidase 1 [DPP1] inhibition), symptoms of patients with Papillon-Lefèvre syndrome (PLS), and findings in nonclinical studies, **periodontal disorders / gingival disorders** were prospectively defined and monitored as Adverse Event of Special Interest (AESI) and categorized as ADR despite overall low frequencies observed in the clinical trials.

In the SOC Neoplasms benign, malignant and unspecified (including cysts and polyps) TEAEs were reported more often in the brensocatib groups, however, no TEAEs in this SOC showed a difference in incidence ≥1.0% between either brensocatib or placebo group. Nevertheless, an association with a broader and numerical higher incidence of malignancies compared to placebo, particularly for skin cancers and with the higher brensocatib dose is noted. The length of exposure limits the strength of the currently available evidence. Malignancies observed among study participants exhibited a latency period of less than one year, a timeframe generally considered too short to support a direct causal relationship

with the investigational drug and not typically consistent with drug-induced malignancy. The clinical data collected to date does not reveal any consistent or characteristic patterns that would indicate the emergence of a clear safety signal. This holds also for skin cancers. Nevertheless, due to the pharmacological mechanism of action of brensocatib and the absence of robust long-term safety data - most notably for the higher 25 mg dose continued pharmacovigilance through post-marketing surveillance will remain essential to detect any potential late-emerging risks. A non - interventional post authorisation safety study (PASS) has been requested and agreed with CHMP The study objectives will be as follows: Evaluation of the long-term safety in patients treated with Brinsupri in the real world setting. The final CSR will be provided in 2034.

Treatment-related TEAEs

Overall, TEAEs were slightly more frequent with brensocatib, especially at 25 mg and occurred in 16.6% of patients on brensocatib 10 mg, 18.4% on 25 mg, and 16.7% on placebo. Related TEAEs were more frequent in the brensocatib group than placebo, and more frequent in the brensocatib 25 mg group than the 10 mg group. The related TEAEs (≥1% in any treatment group) with higher incidence in the brensocatib 25 mg group than the brensocatib 10 mg group and showing dose ordering were headache, hyperkeratosis, dry skin, gingival pain, and fatigue were the most common treatment-related TEAEs belong to the SOC Skin/subcutaneous tissue disorders, followed by the SOCs gastrointestinal and nervous system disorders.

AESIs

In alignment with the clinical presentation of the Papillon-Lefévre syndrome (PLS, a genetic condition caused by loss of function of the gene that encodes DPP1), Hyperkeratosis, Periodontitis/gingivitis and severe infections including pneumonia were prospectively defined and monitored as AESI. This has been supported previously in an EMA Scientific Advice.

Serious Adverse, Fatal and Discontinuation Events

The overall incidence of SAEs was similar across treatment groups with respiratory and infectious events such as bronchiectasis and pneumonia being most common, particularly in the placebo group. COVID-19 was the only SAE reported in more than two participants in either brensocatib group and was more frequent than in placebo. Most SAEs were not considered related to brensocatib. The number of deaths (n= 15) was lower in the brensocatib groups (0.5% for 10 mg and 0.8% for 25 mg) compared to placebo (1.1%). None of the fatal AEs were considered related to study treatment. From the data presented, the overall frequency of TEAEs leading to treatment or study discontinuation was low and similar between groups (<5%/ ≤ 3% in any treatment group).

Subgroup analyses

Subgroup analysis for age, revealed no clinically meaningful differences in TEAE and SAE frequency and pattern among age groups. However, the frequency of related TEAEs was higher in adolescents, the comparison between populations is, however, highly limited by the small sample size of the adolescent population. Further subgroup analyses have been provided considering sex, BMI, race, ethnic origin, BSI and extrinsic factors (e.g., smoking or region) and inhaled steroids (ICS) at baseline. Additional information was provided on patients with hepatically or renally impairment and pregnancy. Overall, dose adjustment is not deemed necessary in either of the analysed subgroups.

Supportive safety data

Assessment of safety data from supportive studies showed general concordance with the safety profile characterized in the primary safety population. The evaluation revealed no new safety signals or clinically significant safety concerns.

DDI

Pharmacokinetic interactions have been described for Brensocatib (e.g. as substrate of CYP3A and Pgp or CYP3A4 induction), observed alterations of the systemic exposure of brensocatib by strong CYP3A inhibitor (clarithromycin), strong CYP3A inducer (rifampin) and strong Pgp inhibitor / moderate CYP3A inhibitor (verapamil), was considered not clinically meaningful. Throughout the clinical trial program, a considerable portion of participants received concomitant medications reflecting standard treatment frequently prescribed in the target population, especially the elderly. No relevant safety signal has become obvious for any of the evaluated comedications.

Laboratory findings and Vital signs

The data provided do not suggest any clinically meaningful trends or changes in clinical laboratory test values, vital signs, ECG or physical examination. No correlation was observed between thiocyanate concentrations and possible metabolite toxicity events. Brensocatib has been comprehensively assessed for potential abuse liability (see Non-clinical AR) and is considered to have no potential for abuse. No clinical evidence of rebound was observed.

6.4.11.1.1. Overall assessment of available safety data

The primary safety evaluation consists of pooled data from 1.326 patients treaed with 10 or 25 mg brensocatib in studies INS1007-201 and -301 and is considered of sufficient size. Data on 52-week exposure support the intended chronic treatment for adults and potentially also for adolescents. While adolescent representation in the safety database is limited, the available data indicate no age-specific safety signals in this population.

Throughout the clinical programme brensocatib was generally well tolerated at the target dose of 25 mg QD. The proposed ADRs headache, gingival disorders, periodontal disorders, hyperkeratosis, rash and dry skin, dermatitis, skin exfoliation, alopecia, upper respiratory tract infection (URTI) and gastroenteritis were all common, predominantly mild or moderate and rarely led to study discontinuation. Most adverse events were manageable or improved/resolved over time even with continuation of brensocatib exposure.

Given its mechanism of action (neutrophil serine protease inhibition), biological plausibility exists for an increased risk for opportunistic infections.

Overall, the data support an adequate safety profile in the treatment of patients with NCFBE.

6.4.11.1.2. Adverse drug reactions (ADRs) in the SmPC

For the following ADRs a causal relationship between the medicinal product and the adverse events is at least a reasonable possibility, based on their comparative incidence in clinical trials.

Table 89: ADRs proposed for inclusion in the SmPC

Nervous system disorders	
Headache	9.2%
Gastrointestinal disorders	
Periodontal disorder	0.8 %
Gingival disorder	1.2 %
Skin and Subcutaneous tissue disorders	
Rash	4.1%
Dry skin	3.0%
Hyperkeratosis*	5.9 %
Dermatitis	4.2%
Skin exfoliation	1.1%
Alopecia	1.2 %
Infections and infestations	
Upper respiratory tract infections	3.9%
Gastroenteritis	1.8%

*Hyperkeratosis includes skin lesion, keratosis pilaris, exfoliative rash, seborrhoeic keratosis.

6.4.11.2. Conclusions on clinical safety

The overall data from the clinical program suggest that brensocatib is well tolerated at long-term treatment with a dose of 25 mg QD. The proposed ADRs were all common, predominantly mild or moderate and rarely led to study discontinuation. Most adverse events were manageable or improved/resolved over time even with continuation of brensocatib exposure.

The safety profile in adolescent population was similar to the adult population.

As potential long-term risks for (opportunistic) infections and malignancies related to acompound targeting proinflammatory cytokine pathways may become apparent only beyond the usual 52-week observation period, respective data should be collected post-approval in the non inverventional study agreed with the applicant and through the post marketing setting.

7. Risk management plan

7.1. Safety specification

7.1.1. Proposed safety specification

The applicant proposed the following summary of safety concerns in the RMP:

Table 90: Summary of safety concerns in the proposed RMP

Summary of safety concerns	
Important identified risks	None
Important potential risks	Embryo-foetal toxicity
Missing information	Use during breast feeding and in pregnancy Long-term safety (in particular the risk for infections and malignancies)

7.1.2. Discussion on proposed safety specification

The above-described safety concerns are considered appropriate. The description and rational for inclusion of the missing information in the RMP is endorsed.

The applicant has discussed the potential risk associated with live attenuated vaccinations and the current label of the SmPC, which states that concomitant use of live attenuated vaccines has not been evaluated and should be avoided in patients receiving brensocatib. During the clinical trials more than 700 patients received an inactivated vaccine without a differential safety profile versus placebo.

For compounds targeting proinflammatory cytokine pathways, there are inherent safety concerns regarding potential long-term effects on critical physiological functions such as host defence, tumour surveillance, and wound healing. These risks may not become apparent within the typical duration of clinical studies and are often captured under the category of "missing information: long-term safety." The applicant was requested to assess whether these concerns are sufficiently addressed within the current "missing information" category, or whether they warrant a separate and more detailed evaluation within the safety specification. The applicant provided data on brensocatib's immunotoxicity profile based on its mechanism of action, structural features, nonclinical and clinical data as well as metabolite considerations. Brensocatib inhibits the neutrophil enzyme DPP1, thereby reducing pro-inflammatory proteases and limiting tissue damage. Evidence from animal models and patients with DPP1 deficiency demonstrates preserved innate and adaptive immune function, and structural analyses confirmed that brensocatib does not resemble known immunomodulatory agents. Clinical trial data did not indicate immunotoxicity or autoimmunity signals; immune-related adverse events were rare, balanced across brensocatib and placebo groups, and assessed as unrelated. Furthermore, no evidence was found for an increased risk of malignancies or opportunistic infections. Based on these findings, the applicant concluded that brensocatib has no immunosuppressive properties, no detrimental impact on antimicrobial immunity and no immunotoxic potential at therapeutic exposures and therefore does not warrant additional immunotoxicity-related safety concerns in the risk management plan. While these results are reassuring, it must be acknowledged that the long-term effects of this class of medicines in humans remain insufficiently characterised. Potential risks relating to infections and malignancies may only emerge after prolonged treatment and cannot be fully excluded on the basis of the available evidence. The PRAC therefore considers it appropriate to refine the definition of "missing information: long-term safety" by explicitly specifying the potential risks of infections and malignancies. This was endorsed by CHMP.

The missing information "Pregnancy and lactation" was renamed to "Pregnancy and Breast feeding". As there were some findings in the pre-clinical data, the applicant was requested to provide additional data and discuss if pregnancy should be included as an important potential risk. The applicant has provided additional studies in the pre-clinical section. Developmental toxicity studies did not reveal major malformations; however, minor skeletal anomalies (bent scapula and wavy ribs) were observed in rat foetuses exposed to brensocatib. The clinical relevance of these findings in the human setting remains uncertain but cannot be excluded. Therefore, the observation of abnormal bone development during

foetal growth is considered an important potential risk and was included in the safety concerns of the RMP under "embryo-foetal toxicity". At the same time the missing information pregnancy was removed from the list of safety concerns. The missing information breast feeding was renamed to "Use during breast feeding" in the safety concerns listed in the RMP.

Although the product is authorised for use in patients aged 12 years and older, the clinical development programme included only a limited number of adolescent participants. The applicant has subsequently provided additional clinical data comprising of 41 adolescent patients, of whom 39 completed treatment. Despite the small sample size, efficacy and safety outcomes in this population were consistent with those observed in adults, supporting a positive benefit–risk profile. Based on these findings, the applicant concluded that no adolescent-specific safety concern requires inclusion in the RMP. This conclusion is considered acceptable.

7.2. Pharmacovigilance plan

7.2.1. Proposed pharmacovigilance plan.

Planned additional pharmacovigilance activities

A post-authorisation safety study is recommended.

Rationale and study objectives:

The PASS is to evaluate long-term safety data of NCFBE patients treated with Brinsupri, in particular the risk for infections and malignancies.

The objective of the study is to evaluate the long-term safety profile of Brinsupri, including the incidence and risk of malignancies and severe infections in the treatment of NCFBE patients in the real-world setting.

Study design:

Single-arm, open-label, observational, non-interventional cohort study on NCFBE patients treated with Brinsupri. Each patient will be followed for up to approximately five years.

Study population:

The target study population consists of NCFBE patients initiating Brinsupri in European countries. The intended sample size is 1,000 participants. The recruitment will be conducted in collaboration with the European Bronchiectasis Registry (EMBARC) investigator network. Safety outcomes will be evaluated by comparison with carefully matched historical or external cohorts, which may be sourced from EMBARC, other high-quality registries, electronic health records, or claims databases.

Milestones:

- Submission of draft protocol: Quarter (Q) 3 2026

- Start of data collection: Q1 2027

- Interim analysis update report: Q4 2031 (when 500 subjects enrolled and having completed 2 years of follow up)

- Final study report: Q4 2034

III.3 Summary Table of additional Pharmacovigilance activities

Table 12: On-going and Planned Additional Pharmacovigilance Activities

Study Status	Summary of objectives	Safety concerns addressed	Milestones	Due dates
Category 1 - Imposed mandatory additional pharmacovigilance activities which are conditions of the marketing authorisation				
None				
Category 2 - Imposed mandatory additional pharmacovigilance activities which are Specific Obligations in the context of a conditional marketing authorisation or a marketing authorisation under exceptional circumstances				
None				
Category 3 - Required additional pharmacovigilance activities				
Long-term safety PASS (study title TBD) Planned	To evaluate long-term safety, in particular risks for infections and malignancies in patients treated with brensocatib in the real-world setting	- Long-term safety - Long-term risk for infections - Long-term risk for malignancies	Submission of draft protocol	Q3 2026
			Start of data collection	Q1 2027
			Interim analysis update report	Q4 2031
			Final study report	Q4 2034

7.2.2. Discussion on the pharmacovigilance plan

7.2.2.1. Routine pharmacovigilance activities

The applicant proposes the following routine pharmacovigilance activities: adverse reactions reporting and signal detection which is endorsed

7.2.2.2. Additional pharmacovigilance activities

The PRAC, having considered the data submitted, is of the opinion that the proposed post-authorisation PhV development plan is sufficient to identify and characterise the risks of the product.

7.2.1. Plans for post-authorisation efficacy studies

Not applicable.

Risk minimisation measures

7.2.2. Proposed risk minimisation measures

The applicant did not propose any additional risk minimisation measures which is acceptable.

7.2.3. Discussion on the risk minimisation measures

7.2.3.1. Routine risk minimisation measures

The PRAC, having considered the data submitted, is of the opinion that the proposed routine risk minimisation measures are sufficient to minimise the risks of the product in the proposed indication.

7.2.3.2. Additional risk minimisation measures

The applicant did not propose any additional risk minimisation measures, which is acceptable.

7.3. Overall conclusion on the Risk Management Plan

CHMP and PRAC consider that the risk management plan version 1.0 is acceptable. The applicant is reminded that in case of a positive opinion, the body of the RMP and Annexes 4 and 6 (as applicable) will be published on the EMA website at the time of the EPAR publication, so considerations should be given on the retention/removal of Protected Personal Data (PPD) and identification of Commercially Confidential Information (CCI) in any updated RMP submitted throughout this procedure.

8. Pharmacovigilance

8.1. Pharmacovigilance system

The CHMP considers that the pharmacovigilance system summary submitted by the applicant fulfils the requirements of Article 8(3) of Directive 2001/83/EC.

8.2 Periodic safety update reports (PSURs) submission requirements

The active substance is not included in the EURD list, and a new entry will be required. The new list of Union reference dates (EURD list) entry uses the European birth date (EBD) or the international birth date (IBD) to determine the forthcoming Data Lock Points. The requirements for submission of periodic safety update reports for this medicinal product are set out in the Annex II, Section C of the CHMP Opinion. The applicant did request an alignment of the PSUR cycle with the IBD. The IBD is 12 August 2025.

9. Product information

9.1. Summary of product characteristics (SmPC)

9.1.1. SmPC section 4.1 justification

The study population was enriched for frequent exacerbators, consistent with the inclusion criteria. While this supports the ability to detect treatment effects, it limits generalizability. The possibility of extrapolating benefit to patients with fewer than two exacerbations is scientifically plausible but currently not supported by direct evidence. The efficacy in the adolescent patient population is demonstrated based on similar pk profile, the limited adolescent clinical results supported with extrapolation from adult clinical data, including extrapolation in patients experiencing more than two exacerbations

9.1.2. SmPC section 5.1 justification

The data presented in Section 5.1 are justified as they demonstrate that brensocatib acts by inhibiting DPP1, resulting in sustained suppression of neutrophil serine protease activity in vivo, which correlates with clinically meaningful reductions in bronchiectasis exacerbations observed in the pivotal ASPEN trial. These findings establish the pharmacodynamic and clinical basis for the 25 mg once-daily dose.

9.2. User consultation

The results of the user consultation with target patient groups on the package leaflet submitted by the applicant show that the package leaflet meets the criteria for readability as set out in the Guideline on the readability of the label and package leaflet of medicinal products for human use.

9.3. Additional monitoring

Pursuant to Article 23(1) of Regulation No (EU) 726/2004, Brinsupri (brensocatib) is included in the additional monitoring list since it contains a new active substance which, on 1 January 2011, was not contained in any medicinal product authorised in the EU.

Therefore, the summary of product characteristics and the package leaflet include a statement that this medicinal product is subject to additional monitoring and will allow quick identification of new safety information. The statement is preceded by an inverted equilateral black triangle.

10. Benefit-risk assessment

10.1. Therapeutic context

10.1.1. Disease or condition, proposed therapeutic indication

Non-cystic fibrosis bronchiectasis (NCFBE) is a chronic, progressive inflammatory lung disease marked by irreversible dilation of bronchi and bronchioles, often leading to airflow obstruction due to abnormal mucus production. The disease is driven by persistent inflammation and/or infection, resulting in airway wall thickening and severe pulmonary dysfunction. Neutrophil activation in the airways leads to the release of neutrophil serine proteases (NSPs), particularly neutrophil elastase (NE), a key mediator in bronchiectasis pathogenesis (Chalmers and Chotirmall, 2018).

NSPs such as NE, proteinase 3 (PR3), and cathepsin G (CatG) are secreted into the extracellular space where they can exceed the capacity of natural inhibitors like alpha-1 antitrypsin and secretory leukoproteinase inhibitor (Sibila et al., 2019), resulting in airway wall damage (Chalmers et al., 2017), mucus overproduction (Voynow et al., 1999), sustained inflammation, and impaired immune cell function, thereby increasing infection risk (Palmer et al., 2018).

Bronchiectasis exacerbations are marked by worsening respiratory symptoms and are linked to poor outcomes, including reduced quality of life, psychological burden, and increased mortality (Brill et al., 2015; Chalmers et al., 2014; Olveira et al., 2013). Patients with NCFBE typically experience 1.4 to 3.9 exacerbations per year (Chalmers et al., 2014; Goeminne PC, 2014). Frequent exacerbations contribute to lung function decline and disease progression (Araujo et al., 2018; Brill et al., 2015; Chalmers et al., 2018; Kapur et al., 2018) and are associated with persistent neutrophilic inflammation (Chalmers et al., 2017; Finch et al., 2019).

The "*vicious vortex*" model describes the interrelated mechanisms of bronchiectasis, including mucociliary dysfunction, chronic infection, persistent inflammation, and structural lung damage. Each element reinforces the others, driving the cycle of disease (Chalmers and Chotirmall, 2018).

Brensocatib is a first-in-class, oral, selective, competitive, and reversible inhibitor of dipeptidyl peptidase 1 (DPP1), which activates NSPs (NE, PR3, CatG) during neutrophil maturation in the bone marrow.

The approved indication is: "Brinsupri is indicated for the treatment of non-cystic fibrosis bronchiectasis (NCFBE) in patients 12 years of age and older with two or more exacerbations in the prior 12 months".

10.1.2. Available therapies and unmet medical need

Bronchiectasis remains challenging to manage, with no approved pharmaceutical treatments and limited therapeutic options. Current management relies mainly on airway clearance, antibiotics, and anti-inflammatory agents, which provide only partial symptom control and do not address the underlying disease mechanisms. Given the central role of neutrophils and neutrophil serine proteases (NSPs) in NCFBE pathogenesis, DPP1 inhibition represents a novel strategy to directly modulate neutrophilic inflammation. This mechanism distinguishes brensocatib from existing symptomatic and anti-infective therapies, none of which specifically target NSP activity, highlighting a significant unmet medical need in this population.

10.2. Main clinical studies

The pivotal study supporting the evaluation of brensocatib for NCFBE is Study INS1007-301 (ASPEN), a Phase 3, multicentre, randomized, double-blind, placebo-controlled trial. A total of 1,767 participants aged 12–85 years with HRCT-confirmed NCFBE and ≥2 pulmonary exacerbations (≥1 for adolescents) requiring physician-prescribed systemic antibiotics in the prior 12 months were randomized 1:1:1 to receive brensocatib 10 mg, brensocatib 25 mg, or placebo once daily for 52 weeks, followed by a 4-week observation period.

Randomisation was stratified by geographic region, baseline *Pseudomonas aeruginosa* colonisation, prior exacerbation frequency, and age group. Adolescents (n=41, 2.4% of the study population) were randomized separately in a 2:2:1 ratio (10 mg QD, 25 mg QD, Placebo QD) without stratification. Participants and investigators were blinded to treatment allocation; active and placebo tablets were identical in appearance.

The study employed an independent Clinical Endpoint Committee (CEC) to adjudicate all suspected pulmonary exacerbations (PEs). All 2,267 investigator-reported events were reviewed; 1,897 (83.7%) met protocol-defined criteria and formed the basis of the primary endpoint - annualized rate of CEC-adjudicated PEs over 52 weeks. Key secondary endpoints included time to first PE, proportion of exacerbation-free participants, lung function (post-bronchodilator FEV1), annualized rate of severe PEs, and changes in QOL-B Respiratory Symptoms scores.

Study completion rates were ~80% overall and balanced across arms; discontinuations were primarily due to subject withdrawal, adverse events, or external circumstances.

10.3. Favourable effects

Brensocatib demonstrated a statistically significant and clinically relevant reduction in the annualized rate of CEC-adjudicated, protocol-defined pulmonary exacerbations (PEs) over a 52-week treatment period. In the ITT population using the On-Study estimand, the annualized PE rate was 1.015 in the brensocatib 10 mg group and 1.036 in the 25 mg group, compared to 1.286 in the placebo group. This corresponds to a rate ratio of 0.789 (95% CI: 0.680 to 0.916) for the 10 mg dose and 0.806 (95% CI: 0.694 to 0.936) for the 25 mg dose. The adjusted p-values were 0.0038 and 0.0048, respectively. These findings were robust across sensitivity analyses, including reference-based multiple imputation (jump-to-reference), and remained consistent under both treatment policy and alternative estimands.

Key secondary endpoints supported these findings. Time to first PE was significantly prolonged with both brensocatib doses compared to placebo, with hazard ratios of 0.813 (95% CI: 0.695 to 0.952) for 10 mg and 0.825 (95% CI: 0.705 to 0.964) for 25 mg. The proportion of participants who remained exacerbation-free during the 52-week treatment period was also higher in the brensocatib arms, with odds ratios of 1.412 and 1.400 versus placebo for 10 mg and 25 mg, respectively.

Further supportive evidence included a modest attenuation of post-bronchodilator FEV_1 decline in the 25 mg group (+36 mL vs placebo; nominal p=0.0120), consistent directional trends in FVC and patient-reported outcomes (QOL-B RSS, BEST), and greater reductions in neutrophil serine protease activity at 25 mg compared with 10 mg. While these endpoints were exploratory or not alpha-controlled, they provide mechanistic and clinical support to the primary results.

The strength of evidence is high, based on a large, global, randomised, placebo-controlled trial with blinded, independent adjudication of all suspected events, balanced protocol deviations, and prospectively applied multiplicity adjustment. Internal consistency across primary and key secondary endpoints further supports the robustness of the observed treatment effect.

Both the 10 mg and 25 mg doses met the primary endpoint, with adjusted p-values of 0.0038 and 0.0048, respectively. Each dose demonstrated statistically significant reductions in annualised PE rates compared with placebo across multiple estimands (On-IP, On-Treatment, and On-Study). Rate ratios ranged from approximately 0.78 to 0.82, with nominal p-values consistently <0.01. Although both doses demonstrated comparable efficacy on PE-related endpoints, the 25 mg dose provided more consistent effects on lung function, NSP biomarkers, and exploratory QoL outcomes, whereas the 10 mg dose showed numerically favourable results across some exacerbation endpoints. The CHMP considered the justification for selecting 25 mg as the recommended dose acceptable.

10.3.1. Uncertainties and limitations about favourable effects

For the FEV1 endpoint, not all randomized participants in the ITT population were included in the analysis. The applicant provided an additional J2R analysis in the full ITT set, which confirmed no effect for 10 mg and a small but statistically significant attenuation of decline of FEV1 for 25 mg dose (+36 mL vs placebo; nominal p=0.0120). While this supports robustness, the magnitude of benefit is modest and of unclear clinical relevance since the MICD for patients with bronchiectasis is currently unknown. QoL improvements did not reach the established meaningful within-patient change (MWPC) threshold, and hierarchical testing was broken; therefore, these findings are regarded as exploratory.

The adolescent subgroup analysis relied on Bayesian borrowing from adults. The CHMP noted insufficient exploration of operating characteristics (e.g. effective sample size by mixing weight). For the adolescent population, extrapolation remains acceptable based on PK similarity, and similarity of the disease, but efficacy conclusions are limited by small numbers of patients studied in the clinical trial (n=41).

Subgroup findings suggest that brensocatib efficacy may be reduced in subpopulations with features suggestive of type 2 inflammation, more severe airflow limitation, or regional differences in clinical practice and pathogen burden. A post hoc interaction for FEV_1 <50% suggested heterogeneity of treatment effect (p = 0.0053 for 25 mg vs placebo). Given the small sample size and exploratory nature, these results should be interpreted cautiously.

10.4. Unfavourable effects

A total of 1.672 subjects has been exposed to brensocatib. The primary safety evaluation consists of data from 1.326 patients treated with 10 or 25 mg brensocatib for 6 to 12 months. Strength of evidence relies on the pooled database of two adequately sized randomised placebo-controlled studies.

Key unfavourable effects were headache (9.2%), hyperkeratosis (5.9%), dermatitis (4.2%), rash (4.1%), upper respiratory tract infections (3.9%), and dry skin (3.0%), which are all considered common ADRs.

Other unfavourable effects of the SOC Skin and subcutaneous tissue disorders and evidence from the clinical presentation of the Papillon-Lefévre syndrome (PLS, a genetic condition caused by loss of function of the gene that encodes DPP1) support the consistency of the dermatological findings.

Studies in animals have shown a potential embryofoeatal risk in newborns/infants. Therefore, brinsupri is not recommended during pregnancy. Further decision on the need to discontinue treatment during breast feeding will be based on individual patient situation as stated in the SmPC section 4.6.

10.4.1. Uncertainties and limitations about unfavourable effects

While the size and duration of the safety dataset (1,326 patients treated up to 52 weeks) are generally adequate for a chronic indication, several limitations remain.

Additional long-term safety data from the open-label EAP are pending, and the EAP dosing scheme is of limited value for the to be marketed strength.

Most TEAEs were mild to moderate, and the overall incidence of SAEs, discontinuations, and fatal events was low and comparable across groups. However, some uncertainties remain on potential risks regarding opportunistic infections or malignancies after long-term treatment. This is addressed in the RMP. Long term safety will be monitored during the post authorisation setting and through a non interventional post authorisation study requested by the CHMP.

The adolescent data is of limited size and should therefore be interpretated with caution. The safety profile will be further characterised in the post marketing setting.

Information on an embryofoetal risk in newborns/infants has been added as important potential risk in the RMP.

10.5. Effects table

Table 91: Effects table for Brinsupri treatment of NCFBE (data cut-off: 24 March 2024)

Effect (short description)	*Treatment* *10 mg*	*25 mg*	*Placebo*	*Uncertainties/ Strength of evidence*
Favourable effects				
Annualized Rate of PEs Number of participants with ≥ 1 exacerbation event, n (%)	292 (50.1%)	288 (50.1%)	324 (57.5%)	Comparison groups: 10 mg vs Placebo, Rate Ratio 0.789, 95% CI (0.680,0.916) p-value 0.0019 25 mg vs Placebo, Rate Ratio 0.806, 95% CI (0.694,0.936), p-value 0.0046
Time to First PE Median Time to First PE (weeks)	49.000	50.714	36.714	Hazard Ratio (95% CI) 10mg 0.813 (0.695,0.952) p-value 0.0100 25mg 0.825 (0.703,0.968), p-value 0.0182
Responder Status for Exacerbation-free Responders, Average of 100 imputed datasets, n (%)	282.9 (48.5)	278.7 (48.5)	226.7 (40.3)	Odds Ratio (95% CI) 10 mg 1.412 (1.105,1.806), p-value 0.0059 25mg 1.400 (1.095,1.792), p-value 0.0074
Change in FEV_1, Number of participants in model, LS mean (SE) (L)	564 -0.050 (0.0093)	551 -0.024 (0.0099)	539 -0.062 (0.0094)	Uncertainty whether the small effect size (as well as general minor worsening in placebo) is clinically relevant
Annualized Rate of Severe PEs Annualized rate (95% CI)	0.137 (0.103,0.182)	0.137 (0.105,0.179)	0.185 (0.142,0.242)	rate was not stat. significant, possibly reflects low frequency of events, so limited power for detecting a difference

Effect (short description)	*Treatment* *10 mg*	*25 mg*	*Placebo*	*Uncertainties/ Strength of evidence*
Change in QOL-B Respiratory Symptoms Domain Score (adults only) Number of participants in model LS mean (SE)	487 6.841 (0.7706)	495 8.575 (0.7556)	486 4.809 (0.7500)	Participants who reached a within-patient MCT ⩾14 points (responders) at Week 52, was numerically higher in the brensocatib 10 mg group and nominally significant in the 25 mg group compared with placebo (28.3% and 28.0% vs 23.8%, respectively). This is considered a more clinically meaningful MCID score.
Unfavourable effects				
Headache	47 / 663 7.1 %	61 / 663 9.2 %	42 / 648 6.5 %	**SoE:** pooled data (24-52w) Studies INS1007-201 and -301
Hyperkeratosis	29 / 663 4.4%	39 / 663 5.9 %	20 / 648 3.1%	**SoE:** pooled data (24-52w) Studies INS1007-201 and -301
Rash	19 / 663 2.9 %	27 / 663 4.1 %	15 / 648 2.3 %	**SoE:** pooled data (24-52w) Studies INS1007-201 and -301
Dry skin	12 / 663 1.8 %	20 / 663 3.0 %	11 / 648 1.7 %	**SoE:** pooled data (24-52w) Studies INS1007-201 and -301
Upper respiratory tract infections	30 /663 4.5%	26 / 663 3.9%	20 / 648 3.1%	**SoE:** pooled data (24-52w) Studies INS1007-201 and -301
Gastroenteritis	10 / 663 1.5%	8 / 663 1.2%	3 / 648 0.5%	**SoE:** pooled data (24-52w) Studies INS1007-201 and -301
Dermatitis (pooled)	20 / 663 3.0%	28 /663 4.2%	15 / 648 2.3%	**SoE:** pooled data (24-52w) Studies INS1007-201 and -301
Skin exfoliation	8 /663 1.2%	7 / 663 1.1%	0	**SoE:** pooled data (24-52w) Studies INS1007-201 and -301
Malignancies (pooled)	8 / 663 1.2%	16 / 663 2.4%	8 / 648 1.2%	**SoE:** pooled data (24-52w) Studies INS1007-201 and -301
Periodontal diseases	2/663 0.3%	5/663 0.8%	1 / 648 0.2%	**SoE:** pooled data (24-52w) Studies INS1007-201 and -301
Gingival disorder	10/663 1.5%	8/663 1.2%	9 / 648 1.4%	**SoE:** pooled data (24-52w) Studies INS1007-201 and -301
Alopecia	7/663 1.1%	8/663 1.2%	3/648 0.5%	**SoE:** pooled data (24-52w) Studies INS1007-201 and -301

Abbreviations: Ref: reference; Unc: uncertainties; SoE: strength of evidence; <sBA: serum bile acids>; <PELD: paediatric end-stage liver disease>; <MELD: model for end-stage liver disease score>; <SBD: surgical biliary diversion>; <OLT: orthotopic liver transplantation>; <PE: primary endpoint>; <SE: secondary endpoint>; <OR: odds ratio>.

*Adjusted p-values for multiplicity calculated using the enhanced mixture-based gatekeeping procedure. Family 1 was tested at two-sided alpha = 0.01 using Truncated Hochberg procedure with a truncation fraction of 0.9. Family 1 served as a gatekeeper for testing secondary endpoints sequentially in the order predefined in the statistical analysis plan.

Family 2 to Family 6 were tested at two-sided alpha=0.05.

10.6. Benefit-risk assessment and discussion

10.6.1. Importance of favourable and unfavourable effects

Brensocatib has demonstrated a statistically significant and clinically meaningful reduction in the rate of CEC-adjudicated, protocol-defined pulmonary exacerbations (PEs) in patients with NCFBE, a population with a high unmet need and no approved disease-modifying therapies. This is based on a single pivotal study (ASPEN) with a supportive study (WILLOW). The findings were consistent across two doses (10 mg and 25 mg), multiple secondary endpoints (e.g. time to first PE, exacerbation-free status), and sensitivity analyses. These favourable effects are supported by the robust design of the pivotal Phase 3 ASPEN study, including stratified randomisation, independent adjudication of events, and pre-specified multiplicity controls. The internal consistency between primary and key secondary endpoints further strengthens confidence in the observed benefits.

Additional favourable effects were observed at the 25 mg dose, including a modest but statistically significant attenuation of post-bronchodilator FEV_1 decline (+36 mL vs placebo; nominal p = 0.0120), reductions in NSP biomarkers, and supportive trends in QOL-B RSS and BEST scores. However, the absolute magnitude of lung function benefit is small, QoL improvements did not reach the established MWPC threshold, and hierarchical testing was broken; therefore, these findings are exploratory. Subgroup results suggest reduced efficacy in patients with FEV_1 <50% or type 2 inflammatory features.

Although both doses met the primary endpoint, the 10 mg dose showed numerically favourable results across some exacerbation outcomes, while the 25 mg dose provided stronger effects on lung function, biomarkers, and exploratory endpoints. Therefore, the applicant's justification for selecting 25 mg was accepted.

The efficacy in adolescent is extrapolated from PK data with limited support from small Bayesian analyses (n=41) and similarities in between the adolescent and adult conditions. Nevertheless, it is considered that enough evidence has been presented to consider that efficacy has been demonstated in this population.

The unfavourable effects of brensocatib, including headache, gingival/periodontal disorders, dermatitis, rash and upper respiratory tract infection were mostly mild to moderate and mechanistically expected. The dermatological findings are consistent with the drug's DPP1 inhibition and are manageable in clinical practice. Serious adverse events, discontinuations, and deaths were infrequent and balanced across treatment groups. Uncertainties concerning a potential for opportunistic infections and the development of malignancies associated with long-term treatment are mentioned in the RMP and recommended to be addressed post-approval. The safety profile in the adolescent population has swhon to be similar to the one of the adult population.

No major safety signals have emerged to date. The observed risks appear tolerable within the context of a chronic progressive disease with no alternative treatment options. Long-term safety effects are recommended to be followed in the post marketing setting to further characterize brensocatib's risk profile.

10.6.2. Balance of benefits and risks

The demonstrated reduction in PE frequency and delay in exacerbation onset represent clinically relevant benefits for a patient population with limited options. These benefits are supported by a large, well-controlled trial and consistent findings across endpoints and estimands. Supportive effects on lung function, biomarkers, and QoL, particularly with the 25 mg dose, add weight to the conclusion of

demonstrated benefit, though their clinical relevance remains modest. The 25 mg dose is supported as the recommended regimen.

The safety profile is generally manageable, with adverse events that are mostly mild, predictable, and consistent with the mechanism of action. Long-term safety uncertainties are considered addressed through continued data collection, post-marketing surveillance, and appropriate risk mitigation strategies in place.

10.6.3. Additional considerations on the benefit-risk balance

Indication

Inclusion criteria for the pivotal ASPEN study required patients to have experienced ≥2 pulmonary exacerbations (≥1 for adolescents) in the 12 months prior to screening, each defined by the need for physician-prescribed systemic antibiotics for signs and symptoms of respiratory infection. This definition directly reflects the proposed indication. During the assessment, the applicant confirmed that the proposed indication is restricted to this studied population and does not seek approval in a broader NCFBE population. While CHMP noted that patients with fewer exacerbations may still suffer substantial disease burden, the applicant's justification to maintain the threshold of ≥2 events is considered acceptable.

10.7. Benefit-risk conclusions

The benefit–risk balance of brensocatib is positive for the treatment of non-cystic fibrosis bronchiectasis (NCFB) in patients 12 years of age and older with two or more exacerbations in the prior 12 months.