Trial Protocol

A Trial of the Safety, Tolerability and Efficacy of Cayston (Aztreonam Lysine) compared to placebo in participants with bronchiectasis.

Trial Acronym	Value of inhaled treatment with Aztreonam lysine in bronchiectasis- VITAL- BE
Sponsor	University of Dundee-NHS Tayside
Sponsor R&D Number	2016RC27
Funder	Gilead Sciences
Chief Investigator	Professor James Chalmers
Clinical Trials Unit	Tayside Clinical Trials Unit
EudraCT Number	2018-001590-24
CTA Number	21726/0289/001-0001
IRAS number	252929
Clinicaltrials.gov Number	NCT03696290
Version Number and Date	VitalBE Protocol v11 27-02-2024

The clinicaltrials.gov record will be updated regularly, and results will be posted on trial completion.

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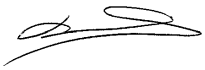
PROTOCOL APPROVAL

A Trial of the Safety, Tolerability and Efficacy of Cayston (Aztreonam Lysine) compared to placebo in participants with bronchiectasis.

EudraCT number: 2018-001590-24

Signatures

The undersigned confirm that the following protocol has been agreed and approved by the Sponsor and that the Chief Investigator agrees to conduct the trial in compliance with this approved protocol and will adhere to the principles outlined in the EU Clinical Trials Directive (2001/20/EC), the EU Clinical Trials Regulation (536/2014) and any subsequent amendments of the Clinical Trials Regulation, the principles of GCP, the Sponsor SOPs, and any other applicable regulatory requirements as may be amended from time to time.

Professor James Chalmers		28-02-2024
Chief Investigator	Signature	Date
Mr Jamie Stobo		28/Feb/2024
Individual Responsible for Statistical Review	Signature	Date

LIST OF ABBREVIATIONS

AE	Adverse Event
ALP	Alkaline Phosphatase
ALT	Alanine Transaminase
AR	Adverse Reaction
BHQ	Bronchiectasis Health Questionnaire
BP	Blood Pressure
BPIS	Brief Participant Information Sheet
BSI	Bronchiectasis Severity Index
CF	Cystic Fibrosis
CI	Chief Investigator
CNORIS	Clinical Negligence and Other Risks Indemnity Scheme
COPD	Chronic Obstructive Pulmonary Disease
CRF	Case Report Form
CRP	C- Reactive Protein
CTA	Clinical Trail Authorisation
CTIMP	Clinical Trial of Investigational Medicinal Product
DMC	Data Monitoring Committee
DMS	Data Management System
DSUR	Development Safety Update Report
ECG	Electrocardiogram
eGFR	Estimated Glomerular Filtration Rate
EMBARC	European Multicentre Bronchiectasis Audit and Research Collaboration
ERS	European Respiratory Society
EudraCT	European Clinical Trials Database
FBC	Full Blood Count
FEV	Forced Expiratory Volume
FVC	Forced Vital Capacity
GCP	Good Clinical Practice
GP	General Practitioner
ICF	Informed Consent Form
ICS	Inhaled Corticosteroids
ISF	Investigator Site File

IMP	Investigational Medicinal Product
LFTs	Liver Function Tests
LPLV	Last Participant Last Visit
MA	Marketing Authorisation
NICE	National Institute for Clinical Excellence
NHS	National Health Service
NHS R&D	National Health Service Research & Development
NRES	National Research Ethics Service
PI	Principal Investigator
PIS	Participant Information Sheet
QC	Quality Control
QOL-B	Quality of life bronchiectasis questionnaire
RCUK	Research Council- UK
REC	Research Ethics Committee
SAR	Serious Adverse Reaction
SGRQ	St George's Respiratory Questionnaire
SHARE	Scottish Health Register
SmPC	Summary of Product Characteristics
SOP	Standard Operating Procedure
SUSAR	Suspected Unexpected Serious Adverse Reaction
TASC	Tayside Medical Sciences Centre
TCTU	Tayside Clinical Trials Unit
TMF	Trial Master File
U&Es	Urea & Electrolytes
UAR	Unexpected Adverse Event
UKCRC	United Kingdom Clinical Research Network
WOCBP	Woman of Childbearing Potential

SUMMARY/SYNOPSIS

Trial Title	A Trial of the Safety, Tolerability and Efficacy of Cayston (Aztreonam Lysine) compared to placebo in participants with bronchiectasis	
Trial Phase & Type	Phase 2 randomised controlled trial	
Trial Design	Multi-centre randomised double-blind placebo controlled parallel group trial with two treatment arms	
Trial Population	Bronchiectasis participants with a history of at least 3 exacerbations in the previous year and the presence of chronic Gram-negative infection in sputum at screening	
Sample Size	77	
Planned Trial Period	2 years	
Clinical phase duration	1 year	
Follow up phase duration	N/A	
Primary	Objectives To evaluate the safety and tolerability of Aztreonam lysine	Outcome Measures Recording of adverse events, serious adverse events and trial treatment withdrawals between groups
	To determine the effect of Aztreonam Lysine on time to first pulmonary exacerbation	Time to first exacerbation
Secondary	Objectives	Outcome Measures
	To determine the effect of Aztreonam lysine on the frequency of exacerbations over 12 months	Frequency of exacerbations
	To determine the effect of Aztreonam lysine on quality of life	St. Georges Respiratory Questionnaire Quality of Life Bronchiectasis Questionnaire Bronchiectasis Health Questionnaire
	To determine the effect of Aztreonam lysine on pulmonary function	*FEV1*
	Bacterial load at the end of the first treatment cycle	*CFU/ml*

Investigational Medicinal Product(s)	Aztreonam lysine (Cayston)
Inclusion Criteria	1. Male or female participants ≥ 18 years of age 2. Clinical diagnosis of Bronchiectasis. 3. Able to give informed consent. 4. CT scan of the chest demonstrating bronchiectasis in 1 or more lobes 5. A history of at least 3 exacerbations in the previous 12 months 6. Bronchiectasis severity index score >4 7. At screening participants should have grown Pseudomonas aeruginosa or other Gram-negative respiratory pathogens in sputum or bronchoalveolar lavage on at least 1 occasion in the previous 12 months. 8. A sputum sample that is culture positive for P. aeruginosa or other Gram-negative respiratory pathogens sent at the screening visit and within 35 days of randomization. Pre-specified eligible organisms include Eschericia coli, Haemophilus influenzae, Moraxella catarrhalis, Klebsiella pneumoniae, Proteus mirabilis, Serratia marcescens, Achromobacter, Enterobacter and Stenotrophomonas maltophilia.
Exclusion Criteria	1. Participants with bronchiectasis due to cystic fibrosis 2. Immunodeficiency requiring replacement immunoglobulin. 3. Active tuberculosis or nontuberculous mycobacterial infection (defined as currently under treatment or requiring treatment in the opinion of the investigator). 4. Recent significant haemoptysis (a volume requiring clinical intervention, within the previous 4 weeks). 5. Treatment with inhaled, systemic or nebulized anti-Pseudomonal antibiotics in the 28 days prior to randomization 6. Oral macrolides which have been taken for a period of less than 3 months prior to the start of the trial 7. Treatment of an exacerbation within 4 weeks of randomization 8. Participants with a primary diagnosis of COPD associated with >20 pack years smoking history. 9. Participants with a history of poorly controlled asthma or a history of bronchospasm with inhaled antibiotics. 10. Pregnant or lactating females. 11. Participants with FEV1 <30% predicted value at screening. 12. . Previous history of hypersensitivity to aztreonam, l-lysine, sodium chloride or lactose monohydrate

	13. Previous history of bronchospasm reported with any inhaled anti-bacterial * 14. Glomerular filtration rate (eGFR) below 30ml/min/1.73m2 or requiring dialysis. This will be determined at screening. 15. Use of any investigational drugs within five times of the elimination half-life after the last trial dose or within 30 days, whichever is longer. 16. Unstable co-morbidities (cardiovascular disease, active malignancy) which in the opinion of the investigator would make participation in the trial not in the participants best interest. 17. Long term oxygen therapy 18. Women of childbearing age or male partners of women of child bearing age and not practicing a method of acceptable birth control (see below) * caution is advised when administering Cayston to patients if they have a history of beta-lactam allergy. Please refer to Cayston SmPC section 4.4

1. INTRODUCTION

Bronchiectasis not due to cystic fibrosis is a chronic inflammatory disease characterised by cough, sputum production and frequent respiratory tract infections. There are currently no licensed therapies for bronchiectasis approved by regulators in the United States or Europe. The disease has a high morbidity, particularly in the presence of chronic P. aeruginosa and other chronic Gram-negative infections. Finch et al demonstrated that the presence of P. aeruginosa infection was associated with a higher frequency of acute exacerbations, worse quality of life and a 3-fold increase in mortality. (Finch et al, Ann ATS 2015). Chronic neutrophilic inflammation is a feature of bronchiectasis and the levels of neutrophilic inflammation predict the risk of future exacerbations (Chalmers et al, AJRCCM 2016). Neutrophilic inflammation is highest in participants with P. aeruginosa and other Gram negative pathogens (Taylor Ann ATS 2015, Chalmers et al, AJRCCM 2016) and inflammation can be suppressed by inhaled antibiotic treatment (Chalmers et al, AJRCCM 2012). There is therefore a strong rationale for the effectiveness of inhaled antibiotic treatment in bronchiectasis.

2. BACKGROUND & RATIONALE

Studies of inhaled antibiotics in bronchiectasis have given mixed results to date. Several open label studies in the late 1980's, testing nebulised β-lactams, demonstrated reduced sputum purulence, sputum volume and improvements in inflammatory markers. (Stockley et al, Clin Ther 1985). In an early phase II double-blind placebo-controlled trial by Barker *et al*. nebulised tobramycin significantly reduced the primary outcome of *P. aeruginosa* bacterial load but was poorly tolerated by some participants (Barker et al, AJRCCM 2000). Subsequently a single centre randomised controlled trial of nebulised gentamicin for 12 months reported significant benefits but was limited by open label design and small sample size (Murray et al, AJRCCM 2011). Haworth et al recruited 144 participants with chronic *P. aeruginosa* infection and randomized participants to nebulised colistin or placebo. The trial narrowly failed to meet its primary endpoint (colistin group 165 days *versus* placebo 111 days; p=0.11). In the secondary endpoints, a large improvement in quality of life using the SGRQ was noted (mean difference −10.5 points; p=0.006) (Haworth et al, AJRCCM 2014).

Aztreonam is an inhaled antibiotic licensed for treatment in cystic fibrosis. Two recent phase III trials in bronchiectasis randomised 266 (AIR-BX1) and 274 (AIR-BX2) participants to Aztreonam 75mg three times daily or placebo over the course of two 28-day treatment cycles (with 28 days off treatment between cycles). The primary outcome was the newly developed Quality of Life Bronchiectasis (QoL-B) questionnaire. Unfortunately the trial failed to meet its primary end-point, with a significant change observed in the QOL-B respiratory symptom score in AIR-BX2 but not in AIR-BX1. Treatment related adverse effects were also increased in the Aztreonam treated participants (Barker et al, Lancet Resp Med 2014)

Reasons for the failure of this trial to meets its endpoint has been the subject of speculation. Likely explanations are that the trial population was quite heterogeneous, with many participants having no history of exacerbations and appearing to have relatively mild disease. The characteristics of the included participants included high rates of pulmonary non-tuberculous mycobacterial disease and COPD. Nadig and Flume compared the characteristics of included participants in this trial to their own population of participants with severe bronchiectasis treated with inhaled antibiotics and identified little correlation, suggesting that the trials included a skewed population that was not representative of real-life clinical practice (Nadig and Flume AJRCCM 2016).

In addition, no dose finding studies were performed in bronchiectasis. The dose of 75mg three times daily was chosen based on efficacy and safety in cystic fibrosis. The rates of

adverse events appear to be higher in bronchiectasis suggesting that doses selected for CF may not be fully appropriate for participants with non-CF bronchiectasis. Whether lower doses may have efficacy and better safety has not been investigated.

There is a need to determine the safety and efficacy of Aztreonam lysine in participants with bronchiectasis and a history of frequent exacerbations.

We hypothesise that Aztreonam lysine will be safe and well tolerated and will reduce the frequency of exacerbations in participants with bronchiectasis and a history of frequent exacerbations. This trial will test Aztreonam lysine compared to placebo. The efficacy and safety of Aztreonam is supported by the evidence for Aztreonam in cystic fibrosis (McCoy AJRCCM 2008) where Aztreonam prolonged the time to first exacerbation by 21 days compared to placebo and improved quality of life. The AIR-BX studies evaluated Aztreonam for inhalation for only 2 treatment cycles. They showed suppression of chronic Gram-negative airway bacterial load but were not designed to evaluate the impact of Aztreonam on the frequency or time to first exacerbation. No attempt to identify the optimal dose was made. The incidence of treatment related adverse effects was increased in AIR-BX1 but was more balanced in AIR-BX2, a trial conducted primarily in European bronchiectasis participants. The reason for this imbalance is unknown.

We conclude from these studies that Aztreonam is effective at suppressing airway bacterial load, but that the efficacy and safety of the treatment is unclear because of the nature of the previous trial design that was focussed on short term improvements in quality of life.

Hypothesis

12 months treatment with Aztreonam lysine for inhalation will be safe and well tolerated and will result in a significant increase in the time to first pulmonary exacerbation in participants with bronchiectasis and a history of frequent exacerbations.

3. TRIAL OBJECTIVES & OUTCOMES

Overall Objectives

To determine if treatment with Aztreonam lysine for inhalation will be safe and well tolerated as compared to placebo (primary safety objective).

To determine if Aztreonam lysine for inhalation will result in a significant increase in time to first pulmonary exacerbation (primary efficacy objective), reduction in the frequency of exacerbations, and additional clinical benefits in participants with bronchiectasis and a history of frequent exacerbations.

Table 1: Primary Objectives and Outcome Measures

Primary Objective:	**Outcome Measure:**	**Time point of outcome measured**
To evaluate the safety and tolerability of Aztreonam lysine	Recording of adverse events, serious adverse events and trial treatment withdrawals between groups	Reported at any time point in the trial.
To determine the effect of Aztreonam Lysine on time to first pulmonary exacerbation	Time to first exacerbation	Single event per participant over 12 months

Table 2: Secondary Objectives and Outcome Measures

Secondary Objective:	**Outcome Measure:**	**Time point of outcome measured**
To determine the effect of Aztreonam lysine on the frequency of exacerbations over 12 months	Frequency of exacerbations	Count data over 12 months
To determine the effect of Aztreonam lysine on quality of life	St. Georges Respiratory Questionnaire Bronchiectasis Health Questionnaire	Continuous variables at 0, 1, 3, 6, and 12 months
	Quality of Life Bronchiectasis Questionnaire	Continuous variables at 0, 1, 2,3,4,5,6,7,8,9,10,11 and 12 months
To determine the effect of Aztreonam lysine on pulmonary function	FEV1	Continuous variables at 0, 6, and 12 months
Change in minimum inhibitory concentration of protocol defined bacteria to aztreonam	MIC	Baseline, 6 months and 12 months
Monitoring of emergent pathogens	Sputum culture results (emergent pathogen is defined as a new organism isolated in sputum culture which was not identified at baseline).	6 months and 12 months

Table 3: Exploratory Objectives and Outcome Measures

Exploratory Objective:	**Outcome Measure:**	**Timepoints measured**
To determine the impact of Aztreonam lysine on the time to first exacerbation, including all clinically treated exacerbations	Time to first exacerbation (protocol defined and non-protocol defined)	Up to 12 months
To determine the impact of compliance on the efficacy of Aztreonam lysine	Compliance recorded and repeat measurement of efficacy end-points in those complying with >80% of doses	Compliance assessments at all trial visits
To store blood and sputum samples for future biomarker and molecular microbiology studies	Biomarker measurement Microbiome studies Antibiotic resistance studies	Future work
To determine the effect of Aztreonam lysine on different exacerbation subtypes	Exacerbations subtyped into those associated with viruses, new bacteria or non-infectious exacerbations	Count data over 12 months

4. TRIAL DESIGN

4.1 TRIAL DESCRIPTION

This is a multi-centre randomised double blind placebo controlled parallel group trial with two treatment arms, with 77 participants planned to be randomized aiming to achieve a total planned evaluable cohort of 70 participants. Participants with bronchiectasis confirmed on CT scanning and a history of at least 3 exacerbations in the previous 12 months will be randomised to treatment with either Aztreonam lysine for inhalation (Cayston) 75mg three times daily, or placebo three times daily. All treatments will be continued for 1 year. As the therapy is administered in 28 days on and 28 days off cycles, this will mean a total of 6 treatment cycles for all participants.

Participant will give informed consent after having adequate time to consider whether they wish to participate in the trial. Participants enrolled into the trial will undergo screening to include a check of inclusion and exclusion criteria, physical examination and medical history. The screening and baseline visits are separate as participants must have sputum samples sent to confirm current infection with Gram-negative airway infection in order to participate in the trial at screening. At the baseline visit participants will be randomized to receive Aztreonam lysine or placebo. Participants will be excluded if commenced on other inhaled anti-pseudomonas antibiotics between screening and baseline visits. Other inhaled anti-pseudomonas antibiotics may be discontinued and the participant re-screened and randomized after 28 days free from inhaled antipseudomonal antibiotics.

Participants will be randomised 2:1 to the trial arms. The design proposed is illustrated below. Following randomisation, participants will be supplied with trial medication appropriate to their specific trial arm. Participants will have the option of collecting/couriered medication on a monthly basis or up to 3 months' supply at a time. Attendances to collect drug will not be classed as extra trial visits. Site staff may contact participants during the trial to remind them to start their next month of treatment, where this does not coincide with a study visit.

Once the participant has been randomised there will be no adjustments to dose of the investigational medications. If possible, no additional treatments should be commenced during the trial period for the treatment of bronchiectasis, excepting the treatment of exacerbations if these are required. However, if clinically indicated, bronchiectasis treatment can be commenced during the trial. This should be recorded in the concomitant medications and the participant should remain on their trial medication. The exception to this is that other inhaled anti-pseudomonas antibiotics which if started during the trial will result in the participant being withdrawn from trial medication.

The treatment period is twelve months in total, with review visits at 4 weeks (telephone call), 12 weeks (telephone call), 24 weeks and 48 weeks (end of trial). (see Appendix 2, Trial Matrix). The treatment duration has been chosen based on results of a US Food and Drug Administration workshop on antibacterial trials in bronchiectasis which concludes that studies of at least 12 months were required to adequately demonstrate effects on exacerbations and to monitor for a sufficiently long period to identify development of antibiotic resistance.

The treatment regime arms are as follows:

Investigational product, dosage and mode of administration

1. Aztreonam lysine for inhalation (Cayston) 75mg three times daily
2. Matched placebo three times daily

The use of these drugs including a placebo is considered to be ethical, as there are no licensed treatments for bronchiectasis or specifically for the treatment of chronic airway *P.*

aeruginosa infection. There is no established standard of care in this participant population and so placebo, with the continuation of all other therapies, is considered to be an acceptable mode of treatment.

Following trial completion, participants should be continued, started or restarted on the appropriate treatment for their bronchiectasis. The treatment for the participant at the end of the trial will be discussed between participant and Investigator and any changes communicated to the GP. The default recommendation will be that participants return to their treatment regime prior to the trial.

4.2 TRIAL FLOWCHART

See Appendix 1 Trial Flow Chart / CONSORT Diagram

4.3 TRIAL MATRIX

See Appendix 2 Trial Matrix

4.4 TRIAL ASSESSMENTS

Trial assessments will be performed according to the schedule in Appendix 2, Trial Matrix. Results from bloods and sputum taken within 5 days of the visit will be valid with the exception of obtaining sputum at screening visit where repeat sputum samples may be provided during the screening period. Where trial assessments identify any clinically significant incidental findings these will be communicated to the participant's GP, with the participant's consent.

The primary endpoint will be time to first pulmonary exacerbation during the 12 month treatment period with a key secondary end-point of the number of exacerbations during the 12 month treatment period.

Participants will be shown how to administer the trial medication at the randomization visit and will receive their first dose under supervision. They will undergo spirometry after trial drug administration to ensure there is no bronchospasm.

Sample storage and transportation will be addressed in the Working Practice Guidelines for Obtaining Trial Samples. Local safety bloods (e.g U&E, LFTs and FBC) will be done at local NHS labs.

4.4.1 Assessment of the exacerbation endpoint

Exacerbations will be defined according to the EMBARC/BRR definition as follows:

1- A deterioration in three or more of the following key symptoms for at least 48 hours
   a. Cough
   b. Sputum volume and/or consistency
   c. Sputum purulence
   d. Breathlessness and/or exercise tolerance
   e. Fatigue and/or malaise
   f. Haemoptysis

AND

2- A clinician determines that a change in bronchiectasis treatment is required.

Events meeting this definition will be regarded as protocol defined exacerbations. The definition includes both exacerbations treated as an outpatient (reported as moderate exacerbations) and those treated with intravenous antibiotics or hospitalization (reported as severe exacerbations). Events receiving antibiotic treatment but not meeting this definition will be regarded as non-protocol defined exacerbations and will be included in a secondary analysis.

Note that exacerbations are not adverse events as they are part of the natural history of the disease. Training will be provided to sites regarding the differentiation of exacerbation symptoms from adverse events relating to treatment. If the participant has an exacerbation during the screening period a trial specific unscheduled visit is not required but the PI will make a clinical decision about whether to see the participant.

If a participant has an exacerbation during the treatment phase, they will be asked to contact the trial team by telephone. Following screening the patients will be provided with a contact number by the study team with clear instructions to contact the team in the event of deteriorating symptoms. The team will arrange for the participant to attend for an unscheduled visit. At the visit the exacerbation event will be assessed following the above criteria. If the clinician decides that antibiotic treatment is required, they will prescribe this medication to the patient. Their next trial visit will be delayed until 7 days after recovery from the exacerbation. The treatment regimen of 28 day (4 weeks) cycle of trial medication will continue despite any changes to their scheduled visit dates, this may require the dispensing of trial medication at the unscheduled visit. If a participant experiences an exacerbation out with the trial team working hours, they will be asked to contact their GP out of hours service. They will be asked to contact the trial team during the next working day and to provide details of any medication prescribed by the GP service and to arrange to attend for an unscheduled visit. If a participant has a home supply of antibiotics and are accustomed to beginning antibiotic therapy at the onset of symptoms then this practice will continue during the trial to enable the patient to maintain control of their therapy. They will be asked to contact the trial team and arrange an unscheduled visit. If a participant is unable to attend for an unscheduled visit as much information as possible will be collected over the phone in order to determine whether the exacerbation is a protocol-defined exacerbation as detailed above. This will be recorded in the CRF.

Unscheduled visits should not be performed if patients have symptoms suggestive of COVID19. If the clinical team determine that a face-to-face unscheduled visit is not appropriate, then those procedures that can be performed remotely (e.g., over the telephone) should be completed.

4.4.2 Standard bacterial culture for screening

P. aeruginosa or Gram-negative bacterial identification during the screening period is required for inclusion. Participants will provide spontaneous sputum samples at screening. These will be processed at local NHS laboratories for identification of bacterial pathogens and antibiotic sensitivity testing, including sensitivity testing for Aztreonam. Samples will be stored at -80C and subsequently transferred to the central laboratory where future testing for resistance to Aztreonam and other studies will be performed. If participants are unable to provide a spontaneous sputum sample at the screening visit, further samples can be submitted for testing during the screening period. The participant will be asked to provide a sample produced at home on the morning of any trial visit which will be used if a second sputum sample is not obtained during the visit. Research staff can send a participant a sputum pot prior to any visit for them to use ahead of the visit.. Ideally this sample should be obtained within 2 hours of the trial visit. Samples collected out with this period will be acceptable if this is the best available.

At the Tayside site rapid sputum PCR using the Biofire PCR system (Biofire Pneumonia Panel) may be used to confirm the presence of protocol defined Gram-negative pathogens. This may allow combined screening and randomization in the same day (the Biofire system takes approximately 1 hour to obtain a result). If there is sufficient sputum an aliquot will still be send for culture and sensitivity testing, but the PCR is sufficient to confirm eligibility. If other sites have the ability to perform rapid sputum PCR they may also carry this out if desired.

Where the screening sputum sample does not identify *P. aeruginosa* or Gram-negative bacterial isolation repeat samples may be obtained during the screening period.

4.4.3 Standard bacterial culture at follow-up visits

Sputum samples from visits, 5 and6 and any unscheduled visits will be obtained at each visit, these will be processed at local NHS laboratories for identification of bacterial pathogens and antibiotic sensitivity testing, including sensitivity testing for Aztreonam. The results of Aztreonam sensitivity will not be made available to the research team to ensure blinding of participants is not compromised. The results of these samples will be assessed by a local/central microbiologist who will inform the PI/delegate of any clinical intervention required.

4.4.4 Nasal swabs and sputum for viruses

At baseline, end of trial and during unscheduled visits participants will have nasal swabs performed as well as having nucleic acid extraction of sputum samples for viral PCR. Swabs will be provided to sites and will be sent for analysis to Professor James Chalmers' laboratory at the University of Dundee.

4.4.5 Symptoms and quality of life

These will be evaluated using the St. George's Respiratory Questionnaire (SGRQ), the Quality of life bronchiectasis questionnaire 3.1 and the Bronchiectasis Health questionnaire (BHQ). Severity of bronchiectasis will be evaluated using the Bronchiectasis Severity Index. These questionnaires will be completed at study visits as outlined in the Study Matrix (appendix 2). Quality of life bronchiectasis questionnaire 3.1 will be completed each month by participants. When this does not coincide with a study visit questionnaires will be posted to participants with a stamped addressed envelope for return to the site. Site staff may contact the participant by telephone to remind them to complete and return the questionnaire.

4.4.6 Spirometry

Forced expiratory volume in 1 second (FEV1) and forced vital capacity (FVC) will be measured. Lung function testing will use ERS standards. The order of spirometry will be as follows:

Visit 1, 5 and 6 and unscheduled visits - post-bronchodilator spirometry only is performed.

Visit 2. Post-bronchodilator spirometry performed. Trial drug is then administered and 15 minutes after completion of administration of trial drug, spirometry is performed again. If there is a >15% reduction in the FEV1 comparing the post-bronchodilator value with the post-trial drug value, then the participant should receive nebulised salbutamol and spirometry repeated. If the FEV1 remains 15% below the baseline post-bronchodilator value then the participant should be withdrawn from the trial and the participant managed by the site according to their clinical judgement.

4.4.7 Research blood and sputum

Blood and sputum will be stored for future biomarker and molecular microbiology studies. This is likely to include biomarker measurement, microbiome studies and antibiotic resistance studies. At the Tayside site, whole blood samples will also be used for extraction of peripheral blood neutrophils for inflammation studies. A maximum of 50ml of blood will be drawn at each visit.

4.5 SAFETY ASSESSMENTS

4.5.1 Laboratory safety assessments

The following laboratory variables will be measured:

Laboratory Safety Variables

Haematology (whole blood- EDTA)	Clinical Chemistry (serum or plasma)
B-Haemoglobin (Hb)	S/P-Creatinine

B-Leukocyte count	S/P-Bilirubin, total
B-Leukocyte differential count (absolute count)	S/P-Alkaline phosphatise (ALP)
B-Platelet count	S/P- Urea
	S/P-Alanine transaminase (ALT)
	S/P-Albumin
	S/P-Potassium
	S/P-Sodium

4.5.2 Physical examination

A detailed physical examination will be performed at screening to exclude participants with co-morbidities or other clinical disorders that would constitute an exclusion from the trial. This will include the following systems:

- Respiratory
- Cardiovascular
- Abdominal
- Neurological
- Dermatological

4.5.3 ECG

A 12-lead ECG will be performed at screening as part of the evaluation to ensure that participants are not enrolled when they have an unstable cardiovascular co-morbidity.

4.5.4 Vital signs

The following will be performed routinely at all trial visits:
Pulse, seated blood pressure, oxygen saturations and body temperature.

4.5.5 Pregnancy Testing

Women of child bearing potential (WOCBP) will have a serum pregnancy test performed pre trial start (at the screening visit) and urine pregnancy tests during the trial at all trial visits. See also notes in the exclusion criteria section regarding contraception and allergy

Under exceptional circumstances (*which may include: in the event of another lockdown, it is felt unsafe for the participant to come to hospital as they should be self-isolating or participant declines visit due to COVID risk*) visits 5 and 6 and any unscheduled visits could be conducted over the telephone. The telephone appointment would include asking about any exacerbations, AE's, changes to con meds, check if any WOCBP is pregnant and the completion of questionnaires only. No other trial procedures would be completed. Spirometry, however, must be performed within the clinical environment at screening and randomisation visits (visits 1 and 2 respectively) to ensure that the first dose of medication is safely tolerated.

4.6 INCIDENTAL FINDINGS

Any incidental findings (IF: previously undiagnosed condition) considered to be clinically significant will be reported to the participant's GP and/or consultant by the CI or Site PI, with the consent of the participant.

5. TRIAL POPULATION

5.1 NUMBER OF PARTICIPANTS

This is a randomised multicentre parallel-group double blind placebo-controlled design with treatment duration of twelve months following screening and randomization. Up to 77participants will be randomised to one of the 2 treatment groups with the aim to have 70 participants completing the trial. Participants will be recruited from approximately 10 NHS Sites in the UK. Participants withdrawn due to bronchospasm after initial drug administration will be replaced.

5.2 INCLUSION CRITERIA

- ≥ 18 years of age
- Able to give informed consent.
- Clinical diagnosis of Bronchiectasis:
- CT scan of the chest demonstrating bronchiectasis in 1 or more lobes
- A history of at least 3 exacerbations in the previous 12 months
- Bronchiectasis severity index score >4
- *Pseudomonas aeruginosa* or other Gram-negative respiratory pathogen detected in sputum or bronchoalveolar lavage on at least 1 occasion in the previous 12 months*.
- A sputum sample that is culture or PCR positive for *P. aeruginosa* or other Gram-negative respiratory pathogens sent at the screening visit and within 35 days of randomization. Pre-specified eligible organisms include *Eschericia coli, Haemophilus influenzae, Moraxella catarrhalis, Klebsiella pneumoniae, Proteus mirabilis, Serratia marcescens, Achromobacter, Enterobacter and Stenotrophomonas maltophilia*

*If a potential participant has not had a sputum sample tested for Pseudomonas aeruginosa or other Gram-negative respiratory pathogens in the previous 12 months then they should be invited for a screening visit, where otherwise potentially eligible. A positive sample at screening or within 35 days prior to randomisation will meet eligibility criteria. Where a participant has had sputum samples tested for Pseudomonas aeruginosa or other Gram-negative respiratory pathogens with only negative results in the previous 12 months they should not be invited for a screening visit.

5.3 EXCLUSION CRITERIA

- Participant has cystic fibrosis.
- Immunodeficiency requiring replacement immunoglobulin.
- Active tuberculosis or nontuberculous mycobacterial infection (defined as currently under treatment or requiring treatment in the opinion of the investigator).
- Recent significant haemoptysis (a volume requiring clinical intervention, within the previous 4 weeks).
- Treatment with inhaled, systemic or nebulized anti-Pseudomonal antibiotics in the 28 days prior to randomization
- Oral macrolides which have been taken for less than 3 months prior to the start of the trial.
- Treatment of an exacerbation and receiving antibiotic treatment within 4 weeks prior to randomization
- Primary diagnosis of COPD associated with >20 pack years smoking history.
- History of poorly controlled asthma or a history of bronchospasm with inhaled antibiotics.
- Pregnant or lactating females.
- Participants with FEV_1 <30% predicted value at screening.
- Previous history of hypersensitivity to aztreonam, l-lysine, sodium chloride or lactose monohydrate.

- Previous history of bronchospasm reported with any inhaled anti-bacterial **
- Glomerular filtration rate (eGFR) below 30ml/min/1.73m^2 or requiring dialysis. This will be determined at screening.
- Use of any investigational drugs within five times of the elimination half-life after the last trial dose or within 30 days, whichever is longer.
- Unstable co-morbidities (cardiovascular disease, active malignancy) which in the opinion of the investigator would make participation in the trial not in the participant's best interest.
- Long term oxygen therapy
- Women of child-bearing age or male partners of women of child-bearing age and not practicing a method of acceptable birth control (see below)

** Caution is advised when administering Cayston to patients if they have a history of beta-lactam allergy. Please refer to Cayston SmPC section 4.4

Individuals who are participating in the follow-up phase of another interventional trial, or who are enrolled in an observational trial, will be co-enrolled where the CIs of each trial agree that it is appropriate.

5.4 CONTRACEPTIVE ADVICE

Women of childbearing potential (WOCBP) must be willing to have pregnancy testing prior to trial entry, prior to administration of trial medication and during trial treatment.

A woman is considered of childbearing potential (WOCBP), i.e. fertile, following menarche and until becoming post-menopausal unless permanently sterile. A postmenopausal state is defined as no menses for 12 months without an alternative medical cause.

In addition, WOCBP, who are sexually active, must be willing to use a form of a medically approved birth control method throughout the treatment period. Elimination half-life from serum is 2.1 hours, and serum concentration following inhalation is 100-fold lower than following injection. Patients will be followed up at trial visits, final trial visit is one month after final IMP dose. Approved birth control method are:

- Combined Oral Contraceptive Pill
- Placement of an intrauterine device – 'coil'
- Barrier methods of contraception: male condom only
- Established use of oral, injected, transdermal or implanted hormonal methods of contraception.
- Male partner sterilisation

Men who are sexually active with female partners of child-bearing potential will also be required to use a form of medically approved birth control method as listed above.

6. PARTICIPANT SELECTION AND ENROLMENT

6.1 IDENTIFYING PARTICIPANTS

Identification of potentially eligible trial participants may make use of any or all of the following:

- From secondary care via contact with participants at specialist respiratory clinics or pulmonary rehabilitation classes. Clinic lists and rehabilitation class participant lists will be reviewed by the PI or delegated member of the clinical care or research teams and medical records checked to identify suitable participants who will then either be approached and given the brief Participant Information Sheet (bPIS) when they attend clinic or class or will be posted an invite letter and bPIS. Appropriate Caldicott approval for this activity where carried out by the research team will be sought. Contact at clinic or

class will be by the PI or delegated member of the clinical care team or local clinician. Postage of invitation letters and bPIS will be carried out by the PI or delegate.

- From local Bronchiectasis databases where participants have given prior consent to be contacted for future research projects, e.g., EMBARC registry, or local registers such as TAYBRIDGE, BRONCH-UK, or similar databases with appropriate approval in other NHS Boards/Trusts as defined locally. Local PI or delegated member of the clinical care or research teams will send out invite letters with bPIS to individuals who may be suitable to take part.
- Recruitment of participants registered via the Scottish Health Research Register (SHARE) (http://www.registerforshare.org/index.php)
- From primary care via the Primary Care Networks. These participants will be sent out an invitation letter and bPIS from the GP practice. GP practices will also be asked to display trial posters and bPIS in their waiting rooms.
- Participant identification Centres (PICs)

When first contact is via letter a bPIS will be sent which gives a general overview of the trial. Participants will be asked to contact the research team if they are interested in the trial. When first contact is in a hospital clinic they will be given a bPIS and will be asked to either return an expression of interest in a stamped addressed envelope or to contact a member of the research team by telephone or email; trial staff may also arrange a convenient time to call the participant. Contact details will be provided on the bPIS.

Should individuals express an interest in taking part in the trial, the PI or delegate will contact the individual and asked if they give permission to check their medical notes. Individuals who returned a reply slip may have provided this permission on the slip in which case further contact with them would not be required prior to accessing their medical notes.

Recruitment may also utilise publicity materials including posters, information leaflets and advertisements.

The local PI will be responsible for recruitment but may delegate to other named individuals within the trial team.

6.2 CONSENTING PARTICIPANTS

Interested individuals will be provided with a full participant information sheet (PIS) before they are scheduled to attend their first research visit. They will be given at least 24 hours to consider their participation further in the trial before a screening visit is arranged by the PI or delegate where participants will be able to ask any questions about the trial. If they decide to participate, written informed consent will be obtained and eligibility by all the criteria confirmed.

Where a participant requests to speak with a physician from the trial team the consent process will not be completed until the participant had spoken to the physician and had all their questions answered to their satisfaction.

The original Informed Consent Form will be filed in the Trial Master File (TMF) or Investigator Site File (ISF) and a copy will be given to the participant and a copy will be filed in the participant's medical notes.

For adults who lose capacity their previous wishes will remain valid unless the protocol changes significantly. If this occurs and further consent is required from a participant who has lost capacity, appropriate person will be asked for their consent. This will be fully documented in the patient's notes.

In all cases the CI or delegate will consult with carers and take note of any signs of objection or distress from the participant – the participant will be withdrawn if they raise objection. Where appropriate the participant will be withdrawn from any further clinical intervention and agreement will be sought from a carer to allow data collection.

The informed consent process will be conducted in compliance with Sponsor SOP

6.3 SCREENING FOR ELIGIBILITY

Participants must have documented bronchiectasis and fulfil the entry criteria for the trial. Participants will be consented prior to screening and a screening log will be maintained.

After consent, all screening assessments, excluding taking blood samples, should be completed in the event of a participant being found to be ineligible on any of the entry criteria. In all cases where participants fail screening for a reason that is modifiable, rescreening will be permitted. Specifically, participants having an exacerbation of bronchiectasis requiring antibiotic administration within 28 days can be rescreened after 28 days have elapsed. Participants with recent use of an investigational drug can be rescreened once the elimination threshold no longer requires exclusion from the trial. Participants with unstable co-morbidities can be rescreened if, in the opinion of the investigator, the co-morbidity becomes stable or resolves such that the participant could be included.

Participants who fail to isolate *P. aeruginosa* or other Gram-negative pathogens at the screening visit may send further sputum samples between screening and randomization, until sputum cultures are positive. Once 35 days has elapsed, however, the participant should be regarded as a screen failure and would require to be rescreened in full.

6.4 INELIGIBLE PARTICIPANTS

Where an individual is found to be ineligible for trial participation, they will be thanked and the reasons for the ineligibility fully explained. Any queries or questions will be answered by an appropriate member of the research team. If ineligibility is related to an IF which is considered to be clinically significant, it will be reported to the participant's GP and/or consultant by the CI or Site PI, with the consent of the individual.

Participants will be asked if they may be contacted to be rescreened if they meet the criteria described above.

6.5 WITHDRAWAL PROCEDURES

Side effects of Aztreonam lysine are likely to be minor and expected in the majority of cases.

In AIR-BX1, dyspnoea, fatigue, pyrexia, chills and respiratory tract congestion were reported more frequently in participants receiving Aztreonam compared to placebo. Few differences were observed in AIR-BX2, with only 1 treatment related serious adverse event.

Bronchospasm is the most well recognised side effect/adverse effect of inhaled antibiotics. Participants will report cough, dyspnoea or wheezing post dose, and this may be associated with a reduction in FEV1. Participants will be told to take their bronchodilator prior to taking a dose of trial medication, as per the instructions in the Cayston patient information leaflet. Sites participating in the trial will be trained in the management of participants receiving inhaled antibiotics and the use of appropriate pre-medication (including pre-medication with bronchodilators) will be used to prevent symptoms associated with inhalation. Sites participating in this trial have experience in managing participants receiving inhalational drugs and will be given training in how to manage symptoms whilst maintaining trial treatment if possible. Symptom management may include temporary discontinuation of the trial drug, which will be allowed in order to try to maintain participants in the trial. The drug may be discontinued for up to a total of 28 days during the year long course of treatment. Withdrawal

from the trial will only be recommended when symptoms are severe, persistent, are determined by the investigator to be treatment related and cannot be managed with appropriate measures.

Post-bronchodilator spirometry will be performed at each face-to-face trial visit. At the randomisation visit participants will additionally receive spirometry after the first dose of trial drug has been administered (section 4.4.5) Withdrawal from the trial will occur if there is a >15% reduction in FEV_1 following initial drug administration that does not respond to bronchodilator treatment. Lung function changes at other time points in the trial would not be an indication for discontinuation of trial treatment unless meeting the above criteria of being associated with severe and persistent symptoms which are treatment related and cannot be managed by the site.

When participants discontinue trial treatment the trial team will ask them if they are willing to remain in the trial and complete the trial assessments.

If participants withdraw from the trial and do not wish to return for trial visits, contact with them will be maintained by the PI, trial manager, research nurse or research assistant for 30 days after the participant's last visit to identify any AEs/SAEs. If withdrawal is due to an AE it will be logged as such on the Adverse Event Log.

Participants are free to withdraw from the trial at any time. The reasons, if known, will be recorded in the medical case notes.

Although a participant is not obliged to give reason(s) for withdrawing prematurely, if the participant appears lost to follow up, the CI will make a reasonable effort to ascertain the reason(s), while fully respecting the individual's rights, and will demonstrate that everything possible was done in an attempt to find any participant lost to follow-up. Those lost to follow-up or withdrawn will be identified and a descriptive analysis of them provided, including the reasons for their loss and its relationship to treatment and outcome.

Details of when IMP must be withdrawn are given in Section 7.1.7

6.5.1 Screening to baseline visit

After consent and eligibility has been confirmed all participants will have sputum samples taken for confirmation of chronic Gram-negative infection. Once results are available and final eligibility is confirmed, participants can be randomized. Training in the use of the inhaled antibiotic will be provided by trial staff. Randomization should take place within 35 days of screening. However, the CI can decide to allow randomisation out with this period and both the decision and the reasoning behind it shall be documented in the participant's notes and CRF. If participants experience an exacerbation of bronchiectasis between the screening to baseline/randomization visit requiring treatment with antibiotics, then they should be withdrawn from the trial and may be rescreened once free from antibiotics for 28 days.

6.6 RANDOMISATION

6.6.1 Randomisation Procedure

After successful completion of screening the exclusion criteria will be checked/re-checked prior to randomization. This will be documented in the participant's medical notes and DMS.

Participants who meet one or more of the exclusion criteria will be withdrawn from the trial. The participant may be rescreened as per section 6.3.

Participants will be randomised by PI or delegate to one of the treatment regimens as noted in Section 4.1.

The PI or delegate will use a centrally controlled web based GCP compliant randomisation system, TRuST, run by the UKCRC registered Tayside Clinical Trials Unit (TCTU). TCTU use

a validated randomisation program and will securely backup both the randomisation seed and the randomisation allocation. Randomisation will be stratified by site, the presence of baseline macrolide treatment and the presence of P. aeruginosa in sputum at screening. The randomisation allocation will be emailed to Site PI, RN and Clinical Trials Pharmacies. The Trial Manager and CI will receive treatment allocation emails for all participants at all sites.

Participants will be prescribed either Aztreonam Lysine (3 doses per day) or placebo (r 3 doses per day) at the randomization visit. Participants will be provided with the Altera nebulizer system at the randomization and will receive their first trial drug dose together with spirometry as described in section 4.4.5.

They will then be asked to take the inhaled therapy as prescribed for 12 months. This includes taking a bronchodilator prior to each dose of inhaled therapy. If a participant requires a bronchodilator for home use these should be prescribed by site staff. Participants withdrawn at randomisation due to bronchospasm would be replaced.

Randomisation will be conducted in compliance with Sponsor SOP

6.6.2 Emergency Unblinding Procedure

Treatment allocation will be carried out by each centre's hospital pharmacy using the randomisation sequence provided by TCTU. TCTU will use an approved web-based randomisation program and will securely backup both the randomisation seed and the randomisation allocation. TCTU will provide each PI with a login to the IWRS, TRuST, for 24-hour emergency unblinding. If unblinding is required, the PI or delegate shall use the web based unblinding system.

In addition, a paper copy of the allocation will be stored securely in NHS Tayside Clinical Trials Pharmacy. Unblinding will only be carried out where a physician considers that it is necessary for clinical safety.

Unblinding will be conducted in compliance with TASC SOP

7. INVESTIGATIONAL MEDICINAL PRODUCTS

7.1 TRIAL DRUG

7.1.1 Trial Drug Identification

	Investigational product	**Dosage, form and strength**
Arm 1	Aztreonam Lysine (Cayston) for inhalation	75mg three times per day
Arm 2	Matched placebo	Three times per day

Treatment is administered in 28 days on and 28 days off cycles.

7.1.2 Trial Drug Manufacturer

Cayston 75mg powder and saline for nebuliser solution

Gilead Medical Information (UK & Eire), 280 High Holborn, London, WC1V 7EE, UK

Marketing authorisation number: EU/1/09/543/001 and EU/1/09/543/002

7.1.3 Storage and Dispensing

Trial medication will be stored at each Site Pharmacy as detailed in the IMP Management Plan. Trial medications will be stored securely at between 2^{o}C-8^{o}C.

7.1.4 Reference Safety Information

The Summary of Product Characteristics (SmPC) will be held in the Pharmacy Site File (PSF), Trial Master File (TMF) and Investigator Site File(s) (ISF). The Reference Safety Information (RSI) is described in Sections 4.8 of the SmPC and is summarised below at

12.3.1. SmPC available at the time of writing are dated 22-04-2021 Cayston 75mg powder and solvent for nebuliser solution.

Any change to the RSI/SmPC for either IMP will be reviewed and if appropriate will results in a substantial amendment.

7.1.5 Dosing Regime

Dosage and regimens are as shown in the table above (Section 7.1.1)

7.1.6 Dose Changes

No changes to dosage or regimen should be made during the course of the trial.

7.1.7 Discontinuation

The investigator may withdraw a patient at any time if it is in the best interest of the patient and treatment continuation would be detrimental to the patients' well-being. In addition, the trial drug will be discontinued in the following circumstances:

- Pregnancy
- Persistent adverse effects such as bronchospasm which are determined to be severe, persistent, treatment related and not responsive to treatment.
- Inability to use the prescribed inhaled therapy.
- Withdrawal from the trial will occur if there is a >15% reduction in FEV_1 following initial drug administration that does not respond to bronchodilator treatment. Lung function changes at other time points in the trial would not be an indication for discontinuation of trial treatment unless meeting the above criteria of being associated with severe and persistent symptoms which are treatment related and cannot be managed by the site.
- If an allergic reaction to trial drug occurs the trial drug will be stopped, and treatment will be initiated as appropriate. The occurrence of rash may be indicative of an allergic reaction to aztreonam.

Where the trial drug is discontinued the participant will be encouraged to complete the trial visits and assessments as per the protocol to allow for an intention to treat analysis but will be censored in the per-protocol analyses.

7.1.8 CONTINUATION OF DRUG FOLLOWING THE END OF TRIAL

Following the end of trial, participants should be continued, started or restarted on the appropriate treatment for their bronchiectasis. No provision for continuation of trial medications will be made by the trial team or Sponsor.

7.1.9 Overdose

Adverse reactions specifically associated with overdose of Cayston have not been identified. Since the mean plasma concentration of Aztreonam following administration of Cayston (75 mg) is approximately 0.6 µg/ml, compared to serum levels of 54 µg/ml following administration of Aztreonam for injection (500 mg), no safety issues associated with Aztreonam overdose are anticipated. If participants report an overdose the medication will be stopped, and the participant will be monitored until resolution of any symptoms. Participants shall be reviewed by the investigator and a medical decision taken as to whether it is safe for the participant to continue in the trial.

7.2 COMPARATOR

7.2.1 Comparator Identification

Lactose Monohydrate placebo for inhalation (5mg/vial), Material Status Notification 16581 and saline for nebuliser solution.

7.2.2 Comparator Manufacturer

Gilead San Dimas

7.2.3 Storage and Dispensing

Storage and dispensing arrangements will be the same as for active drug.

7.2.4 Reference Safety Information

The simplified IMP Dossier (IMPD) will be held in the Trial Master File – specifically in the Pharmacy Site File (PSF) and Investigator Site File (ISF). Any changes to the IMPD for the placebo will be reviewed and if appropriate will result in a substantial amendment.

7.2.5 Dosing Regime

As detailed in section 7.1.1.

7.2.6 Dose Changes

There will be no dose changes.

7.2.7 Withdrawal

As detailed in section 7.1.7

7.2.8 Overdose

Overdose of lactose monohydrate has no known ill effects and requires no specific action. If the participant feels unwell after taking greater than the required dose, symptomatic treatment shall be advised by the research team, with referral to the participants GP or hospital in case of persisting or worsening symptoms.

8. ACCOUNTABILITY PROCEDURES

All IMP will be supplied by a qualified clinical trials medication supplier to the Clinical Trial Pharmacies at sites. Trial medication must be received by a delegated person at the trial Site Clinical Trial Pharmacy, handled and stored safely and properly, and kept in a secured location as detailed in the IMP Management Plan. Pharmacy staff will be responsible for dispensing. All trial clinical supplies are to be dispensed only in accordance with the protocol.

The Investigator or delegated Clinical Trial Pharmacy staff will maintain an accurate record of the receipt and dispensing of IMPs in a drug accountability log. Monitoring of drug accountability will be performed during site visits and at the completion of the trial. Participants will be asked to return all unused and used medications and packaging at each trial visit and at the end of the trial or at the time of discontinuation of treatment.

On return of the medicines, the Clinical Trial Pharmacy staff will perform a check of returns and this will be recorded on the drug accountability log. Unused treatment will be disposed of by the Clinical Trial Pharmacy as per local SOP. Non returned IMPs will be recorded by clinical trials pharmacy staff as per the IMP management plan.

8.1 ACCOUNTABILITY FOR ALL TRIAL IMPS WILL BE IN COMPLIANCE WITH SPONSOR SOPCOMPLIANCE

Trial drug compliance will be assessed by trial personnel at the one month, three month, six month and twelve months (end of trial) visits. The Investigator or delegate will collect used and unused medication and packaging from the participant. Trial drug compliance will be assessed from the information provided by the participant and the above medication checks.

8.2 OTHER MEDICATIONS

8.2.1 Permitted Medications

During the trial it is anticipated that the participant should continue on their usual treatment for bronchiectasis, excluding inhaled anti-Pseudomonal antibiotics.

8.2.2 Restricted Medications

Restricted medications are those that, in the opinion of the Investigators, are likely to result in changes to the time to first pulmonary exacerbation or the frequency of exacerbations. Restricted medications therefore include new initiation of corticosteroids (inhaled and systemic), mucoactive or long-term antibiotics drugs during the treatment period.

Ideally, participants should not be commenced on treatment with these medications during the trial however, if for clinical reasons, a participant is commenced on a restricted medication during the trial, this will be noted in the CRF and the participant records. The patient should continue in the trial and continue to receive trial medication, however, recording this will allow an analysis to take into account any potential effects of these interventions.

Participants should not receive other inhaled or oral long-term antibiotics including macrolide treatment during the trial. If this is required clinically, the participant should continue in the trial and a decision whether the participant should continue the trial medication will be made by the CI.. Patients may be included in the trial if they are receiving these medicines at baseline with the exception of those listed in the exclusion criteria (inhaled or systemic antipseudomonal antibiotics in the prior 28 days and macrolides commenced within the previous 3 months).

8.2.3 Concomitant Medications

Details of all concomitant medications will be recorded in the participant's medical notes or on the concomitant-medications log which should then be saved in the participants medical notes.

9. DATA COLLECTION & MANAGEMENT

9.1 DATA COLLECTION

The Investigator will maintain source documents for each participant in the trial, consisting of case and visit notes (hospital or clinic medical records) containing demographic and medical information, laboratory data, electrocardiograms, trial questionnaires and the results of any other tests or assessments. All trial data relevant to a participant's general medical history will be recorded in the case note. Essential information regarding trial participation will also be recorded in the case note.

The CI may delegate eCRF completion but is responsible for completeness, plausibility, and consistency of the eCRF. Any queries will be resolved by the CI or delegated member of the trial team.

9.2 DATA MANAGEMENT SYSTEM

Data management will be conducted in compliance with Sponsor SOP

A data management system (DMS) will be provided by TCTU using CASTOR. The trial system will be based on the protocol for the trial and individual requirements of the Investigators. Development and validation of the trial database and QC and extraction of data will be done according to TCTU procedures. Extracts for analysis will be based on the dummy data tables provided by the trial team. Core laboratory data will be provided prior to data analysis.

The eCRF will not collect more information than is required to meet the aims of the trial and to ensure the eligibility and safety of the participant. All electronic data will be stored on University of Dundee computers which are password protected and have disaster recovery systems in place.

Database lock will be conducted in compliance with Sponsor SOP.

Note:

At the end of 2019, TCTU made a strategic decision to change the DMS used for trials from OC to Castor, based on the improved functionality of Castor. Initially any trials started in OC would continue in OC until database lock, but all new trials would be developed in Castor, VitalBE used OC as its DMS. However, as recruitment to VitalBE was paused during the COVID-19 pandemic and re-starting the trial involved several changes to the protocol which would mean changes to the existing OC database it was decided that data from new participants would be entered into a new Castor database. The options for how best to amalgamate the current data held in OC and future data to be held in Castor were discussed. Feedback from the Health Informatics Centre, UoD, was that they can't guarantee that if we archive an OC instance, we'll be able to fire it up again. Based on this, and the feedback from all parties, including the TCTU statisticians, the following proposal was adopted:

a) The VitalBE OC instance was locked according to TCTU processes. This included data cleaning and query resolution, a final database audit and a pre-database lock meeting to complete the Database Lock Checklist. A full extract from the database was taken and is held on the TCTU S drive. The OC instance is held on the current server. Should there be any stability issues, we have locked the database as per TCTU WIs and have a full extract. Should we need to unlock the OC instance, we can do so from this server, rather than the server holding the archived versions.
b) The OC instance will not be archived until the trial is archived as a whole, as per TCTU WIs and TASC SOPs.
c) No data will be transferred from OC into the Castor DMS until/unless advised to do so by the trial statistician.
d) Trial analysis will be completed using both data sets, this will be described in the Statistical Analysis Plan.

10. STATISTICS AND DATA ANALYSIS

10.1 SAMPLE SIZE CALCULATION

The primary outcome of the trial is to establish safety and as such no formal power calculation for this outcome has been performed. Rates of treatment related adverse events in AIRBX1 were 40% vs 28% and for AIR-BX2 were 33% vs 18%. We anticipate in this participant population that rates of treatment related adverse events will be 15-20% and will be equal between groups. Rates of discontinuation were 22% vs 6% in AIRBX1 and 10% vs 5% in AIRBX2. We anticipate 10% discontinuation rates in this trial as were observed in a previous trial of nebulised gentamicin (Murray et al, 2011).

The trial by Murray et al observed a significant improvement in time to first exacerbation (120 days vs 61.5 days, HR 0.51, p=0.02 with 27 active and 30 placebo treated participants). Our sample size of 30 participants per group is therefore based on this experience. Participants withdrawn due to bronchospasm after initial drug administration will be replaced.

For frequency of exacerbation using a Poisson distribution with a mean rate of 3 per participant per year in the placebo group (based on the baseline 3 per year as an inclusion criteria), a 50% reduction in the rate with active treatment group the mean exacerbation rate is set at 1.5 per participant per year. With 30 participants per treatment group followed up for 1 year, this reduction is detectable with 90% power, given a test size alpha of 0.05 (2-sided).

In summary, this is intended firstly to establish safety, and secondly is powered to show large effects in terms of efficacy similar to those observed in a previous trial of nebulised gentamicin.

10.2 PROPOSED ANALYSES

Analysis will be based on the intention-to-treat principle as outlined in the ICH E9 'Statistical Principles for Clinical Trials'.

The primary analysis will be a comparison of safety between groups, comparing the rates of adverse events, serious adverse events and trial withdrawals between groups. Statistical comparisons will use standard bivariate statistics.

For secondary outcomes the time to first pulmonary exacerbation utilising survival analysis. The key secondary outcome of frequency of exacerbations will utilise Poisson regression.

Continuous outcomes such as pulmonary function will utilise methods such as linear regression. Pre-specified subgroup analyses will be completed by fitting the appropriate interaction term in the regression model and - if significant - outcomes will be presented separately by level of subgroup. A subgroup analysis will be performed in participants with and without prior macrolide use. The trial will be event driven to ensure sufficient exacerbation events for statistical power for the primary outcome.

A statistical analysis plan (SAP) will be prepared for analysis of primary and secondary outcomes and will include a plan for handling missing data.

10.2.1 Interim Analysis

No interim analysis is planned.

10.3 MISSING DATA

The primary analysis will be based on the intention-to-treat principle. The extent of missing data will be examined and the reason for drop-out ascertained. Multiple imputation may be used to impute missing values if necessary and where assumptions for missing at random (MAR) data are met. Where imputation is used, a sensitivity analysis will be conducted considering only cases without missing data. Complete case analysis where missing participants are excluded will be carried out as a secondary analysis.

With regards to participant medication, we will record this at baseline and trial visits and will perform a secondary analysis adjusting for changes in trial medications.

10.4 TRANSFER OF DATA

Delegated research staff will enter the data required by the protocol into the eCRFs following training in the definitions and methods used in completing the eCRF. On completion of data collection the Investigator must certify that the data entered into the eCRFs are complete and accurate, and fully documented in the participant's medical notes

Data verification and cleaning will be performed as per TCTU local procedures and detailed in the Data Management Plan.

Data preservation and sharing will be in accordance with established procedures at the University of Dundee. General laboratory data methods and results will be documented in laboratory notebooks and then analysed and written up for publication for dissemination to the scientific community. All electronic data will be stored on University of Dundee computers which are password protected and have disaster recovery systems in place. Transfer of data will be via a secure web-based case report form. All data and laboratory notebooks will be retained for at least ten years, in accordance with general RCUK guidelines.

11. ADVERSE EVENTS

11.1 DEFINITIONS

Safety reporting will be conducted in compliance with Sponsor SOP Adverse Event (AE)	Any untoward medical occurrence in a clinical trial participant administered a medicinal product and which does not necessarily have a causal relationship with this product.
Adverse Reaction (AR)	An untoward and unintended response in a participant to an investigational medicinal product which is related to any dose administered to that participant.
Serious Adverse Event (SAE)	A serious adverse event is any untoward medical occurrence that: • results in death • is life threatening • requires hospitalisation or prolongation of existing hospitalisation • results in persistent or significant disability or incapacity • is a congenital anomaly or birth defect • Or is otherwise considered serious
Serious Adverse Reaction (SAR)	An adverse reaction, defined in the RSI, which is serious, as defined above.
Suspected Unexpected Serious Adverse Reaction (SUSAR)	A serious adverse reaction, the nature and severity of which is not consistent with the information about the medicinal product in question set out in the Reference Safety Information.

11.2 RECORDING AND REPORTING AE

All AEs will be recorded on the AE Log in the CRF and will be assessed for severity and causality by the CI or PI. AEs will be recorded from the time a participant consents to join the trial until the participant's last trial visit. An AE may be classified as a serious adverse event (SAE) or adverse reaction (AR). An initial assessment of expectedness for SAEs will be conducted by the Investigator.

The Investigator will make a clinical judgment as to whether or not an AE is of sufficient severity to require the participant's discontinuation of treatment. A participant may also voluntarily discontinuation of treatment due to what he or she perceives as an intolerable AE. If either of these occurs, the participant should be given appropriate care under medical supervision until symptoms cease, or the condition becomes stable. If the participant wishes to withdraw from the trial they should be offered an end of trial assessment.

Participants with unresolved AEs/SAEs at end of trial will be followed up until 30 days after participant's last visit. Suspected Unexpected Serious Adverse Reaction (SUSARS) will be followed until resolution.

The CI, PI or delegate will ask about the occurrence of AEs and hospitalisations at every visit during the trial. Serious AEs (SAEs) will be submitted on an SAE form to the Sponsor

Pharmacovigilance Section Tay.pharmacovigilance@nhs.scot within 24 hours of becoming aware of the SAE. Site PIs will also notify the CI when submitting an SAE.

Worsening of bronchiectasis during the trial will not be classed as an AE but is defined as an outcome. Hospitalisations resulting from worsening of bronchiectasis are common events for patients with bronchiectasis and therefore will be recorded but not reported to sponsor. The exception to this is when, in the opinion of the investigator, there is causal relationship between the trial drug and the hospitalisation for bronchiectasis. Pre-specified outcome(s) will not be classed as an AE but as an outcome. Elective admissions and hospitalisations for treatment planned prior to randomisation will not be considered as an AE. However, any AEs occurring during such hospitalisations will be recorded.

Safety information will be reported to Gilead as per the contract.

The evaluation of expectedness will be made based on the knowledge of the reaction and the relevant safety information (RSI) in the SmPC (see section 7), as described in Sponsor SOP **The Sponsor will make the definitive assessment on expectedness for the purposes of SUSAR reporting.**

11.3 REGULATORY REPORTING REQUIREMENTS

The Sponsor is responsible for reporting SUSARs to the UK competent authority, the MHRA, and the Research Ethics Committee (REC). Fatal or life threatening SUSARs will be reported within 7 days and non-fatal and non-life threatening SUSARs within 15 days.
ANNUAL REPORTING REQUIREMENTS

Annual reporting will be conducted in compliance with Sponsor SOP.

The following reports will be submitted each year as a condition of the regulatory authorisation to undertake a clinical trial or as a condition of a favourable opinion from a REC.

The Development Safety Update Report (DSUR) will be prepared jointly by the Sponsor Pharmacovigilance Section and CI and submitted by the Sponsor to the MHRA on the anniversary of date of Clinical Trial Authorisation (CTA

The DSUR and reports of SUSARs in the UK, with an HRA CTIMP Safety Report Form, will be sent to REC by the Sponsor Pharmacovigilance Section. Any other safety reports, for example, reports of a data monitoring committee (DMC), will be sent by the CI to REC, with a Safety Report Form, and to the Sponsor.

An HRA Annual Progress Report for CTIMPs will be prepared and submitted by the CI to REC, and copied to the Sponsor, on the anniversary date of the REC favourable opinion.

11.4 URGENT SAFETY MEASURES

The CI or other trial physician will take appropriate immediate urgent safety measures in order to protect the participants against any immediate hazard to their health or safety. The MHRA, REC and Sponsor will be notified in writing within three days.

12. PREGNANCY

Pregnancy is not considered an AE or SAE, unless there is a congenital abnormality or birth defect. Any unexpected pregnancy occurring during the trial and the outcome of the pregnancy, will be recorded on a TASC Pregnancy Notification Form and submitted to the Sponsor Pharmacovigilance Section Tay.pharmacovigilance@nhs.scot within 24 hours of becoming aware of the pregnancy. The pregnancy will be followed up until the end of the pregnancy. If the trial participant is a male, informed consent for follow up will be sought from his female partner.

Pregnancy will result in discontinuation of trial treatment.

13. TRIAL MANAGEMENT

13.1 TRIAL MANAGEMENT GROUP

The trial will be coordinated by a Trial Management Group, consisting of the grant holders, including the CI, co-applicants (JSE and PF) and the research assistant, trial manager and research nurse where appropriate. Details will be held in the TMF.

The trial manager/trial co-ordinator will oversee the trial on a day-to-day basis and will be accountable to the CI. The CI is responsible for the overall conduct of the trial and will be responsible for checking the CRFs for completeness, accuracy and consistency. Any queries will be resolved by a delegated member of the trial team.

A Delegation Log will be prepared, detailing the responsibilities of each member of staff working on the trial at each Site.

The functions of the Trial Steering Committee will be undertaken by the Trial Management Group. No independent Trial Steering Committee will be convened for this trial.

13.2 DATA MONITORING COMMITTEE

A DMC will be established to oversee the safety of trial participants. The terms of reference of the DMC are detailed in the TMF. Minutes of the DMC will be maintained in the TMF.

13.3 INSPECTION OF RECORDS

The CI, PIs and all institutions involved in the trial will permit trial related monitoring, audits, REC review, and regulatory inspection. In the event of an audit, the CI will allow the Sponsor, representatives of the Sponsor or regulatory authorities direct access to all trial records and source documentation.

13.4 RISK

13.4.1 Potential Risk

Potential risks to participants are described below.

Bronchospasm has been extensively discussed in this document and is the major predictable adverse effect of inhaled antibiotics. The previous AIR-BX studies identified an increase in respiratory symptoms such as dyspnoea, fatigue, pyrexia, chills and respiratory tract congestion in participants receiving Aztreonam compared to placebo.

Risks associated with trial outcome procedures:

Venepuncture: carries the risk of bruising and discomfort but will be undertaken by experienced staff with access to additional staff if required.

Spirometry: This is associated with some discomfort at full expiration but is minor and most participants with bronchiectasis are experienced in performing spirometry and tolerate it well. Spirometry will be conducted by staff trained in the performance of spirometry to Association for Respiratory Technology and Physiology guidelines (ARTP) guideline standards.

Risks associated with antibiotic resistance and emergence of new pathogens are addressed below.

All associated risks are well understood and have established procedures for management. Careful monitoring of participants throughout the trial with dedicated care and adverse event monitoring will be undertaken as required. Participants will receive a card containing all trial staff contact details to enable participants to contact staff with any concerns.

Risks associated with trial IMPs.

Cayston/Aztreonam lysine: This is a widely used treatment in cystic fibrosis with an acceptable safety profile. Safety will be ensured by excluding participants with a contraindication to inhaled antibiotics or those with a history of bronchospasm.

Participants will receive the package inserts detailing the medication side effects. The side effects of the IMP shall be discussed with the participant prior to consent being taken.

13.4.2 Antibiotic resistance and emergent pathogens

Sputum cultures and testing for antibiotic resistance will be performed at baseline, 6 months after starting treatment and 12 months (completion of treatment) in this study (To clarify baseline antibiotic resistance testing here refers to resistance testing being performed on the screening sputum sample that grows the bug for inclusion. Repeated sputum samples may be required before the positive sample is provided within a 35-day period between visits 1 and 2). The objective of this is to test for the emergence of antibiotic resistance and any emergent pathogens not present at baseline. Patients attending for unscheduled visits will also have sputum culture and antibiotic resistance testing performed to determine if any adverse events or exacerbations are associated with antibiotic resistant or emergent pathogens. This is consistent with the monitoring approaches for resistance and emergent pathogens which have been conducted in other inhaled antibiotic trials in the UK and internationally (Murray et al AJRCCM 2011, Haworth et al AJRCCM 2014).

Antibiotic resistance is likely to be observed in some individuals in this trial but the risk to patient safety is considered to be low. Treatment with all inhaled antibiotics is associated with an increase in the minimum inhibitory concentrations (MICs) of microorganisms isolated from sputum (Kidd et al J Cyst Fib 2018). In some cases, these changes to MICs will result in the organism exceeding the laboratory breakpoint to define a resistant organism and the organism will be classified as resistant (Kidd et al J Cyst Fib 2018). It is important to be aware that the breakpoints in MIC used to define resistance are determined based on the systemic i.e oral or intravenous concentrations of antibiotics used for treatment of infections. The concentrations of antibiotics administered via inhalation are substantially higher than concentrations given via the oral or intravenous route and so will typically be given at levels that are able to overcome the MIC even in “resistant” microorganisms.

Inhaled antimicrobials have been used for more than 20 years in the management of cystic fibrosis lung infections. Although increases in MICs have been observed with all inhaled antibiotics, this has not translated into loss of clinical efficacy of either inhaled or systemic antibiotics. Tobramycin is the most frequently used inhaled antibiotic in CF and tobramycin is still administered systemically for the treatment of exacerbations in these patients without evidence of treatment failure. Long term treatment with aztreonam in cystic fibrosis patients is associated with small increases in MICs that did not affect patient safety or clinical efficacy (Oermann et al J Antimicrob Chemother 2011). A systematic review by Somayaji et al recently concluded that antimicrobial susceptibility testing did not impact on clinical outcomes of antibiotic treatment in individuals with cystic fibrosis (Somayaji J Cyst Fibros 2019). In the most recent large study of inhaled antibiotics in bronchiectasis of inhaled liposomal ciprofloxacin, increases in MICs were observed with treatment after the first treatment cycle which remained elevated through a 1-year treatment period (Haworth et al Lancet Respir Med 2019). MICs reduce during the off-treatment period because acquisition of resistance is associated with a fitness cost for organisms (i.e. resistant organisms are often less virulent and able to persist) leading them to be out-competed by sensitive organisms after the antibiotic selection pressure is removed. (Figure 1)

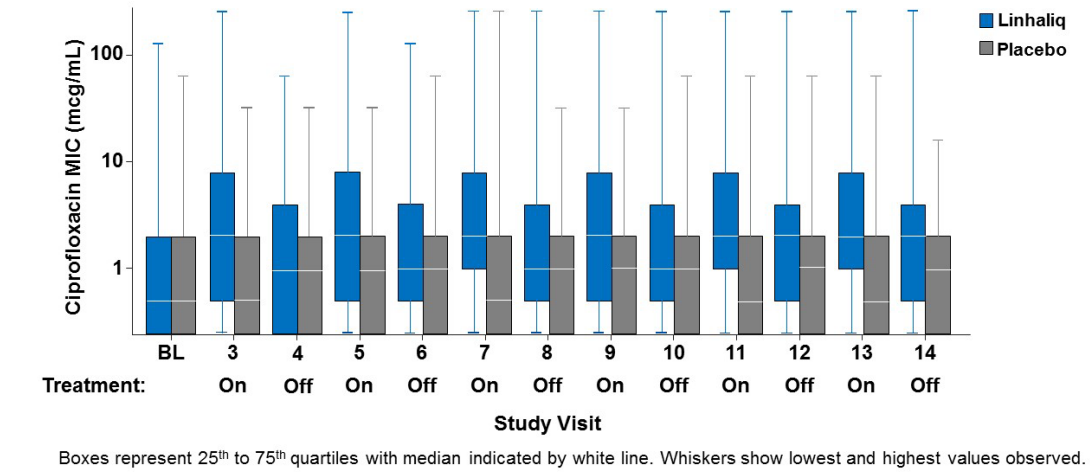


Figure 1. Ciprofloxacin MICs during a 48-week trial of inhaled liposomal ciprofloxacin (linhaliq) vs placebo in bronchiectasis patients with *P. aeruginosa* infection.

Aztreonam is not a first line treatment for exacerbations of bronchiectasis in the UK, where most frequently patients are treated with other antibiotics such as colistin, tobramycin or gentamicin by inhalation. Development of aztreonam resistance is therefore not expected to be a significant clinical risk. Cross-resistance to other antibiotics is a potential risk of all antibiotic resistance mechanisms and this will be monitored throughout the trial by testing MICs against a panel of relevant clinically used antibiotics.

The DMC for the trial includes a microbiologist and will monitor the unblinded results of antimicrobial susceptibility testing and the culture results to identify issues with antibiotic resistance and/or emergent pathogens. The DMC has the ability to recommend termination of the study if they identify an issue which endangers the safety of participants.

13.4.3 Overall risk and benefit ratio of the trial

The overall risk of participating in the trial is considered to be low. Aztreonam lysine is widely used in the treatment of cystic fibrosis in the UK and internationally. Aztreonam was associated with an increase in adverse events leading to trial drug discontinuation in the AIR-BX1 study, but in a population that was not representative of those receiving inhaled antibiotics in clinical practice. Inhaled antibiotics are widely used in bronchiectasis and are considered to be safe. The trial is likely to observe an increase in antibiotic resistance defined by increasing MICs in common with all antibiotic trials. The hypothesis of the trial is that the potential benefit of reducing antibiotic exposure through reducing exacerbations and improving symptoms and quality of life will outweigh concerns over induction of resistance. The trial will provide important scientific benefits by understanding more about the impact of these therapies on the risk of exacerbations.

13.4.4 Risk Assessment & Monitoring

A trial risk assessment was carried out by the Sponsor prior to Sponsorship approval being granted. The Sponsor has determined the appropriate extent and nature of monitoring for the trial and will appoint appropriately qualified and trained monitors. A trial monitoring plan will be prepared and reviewed regularly throughout the trial.

A data safety monitoring board will be convened to review unblinded data from the trial. The independent DSMB will review adverse events, serious adverse events and the results of sputum microbiology including the emergence of resistance and emergent pathogens. The

study stopping criteria will be any serious safety concerns that indicate a potential hazard to health caused by treatment with the IMP.

14. TRIAL CONDUCT & RESPONSIBILITIES

The CI will be responsible for the conduct of the trial. Site delegate(s) will oversee the trial and will be accountable to the CI. A trial-specific Delegation of Duties & Signature Log will be prepared for each Site, detailing the duties of each member of staff working on the trial.

14.1 APPROVALS

The trial will be conducted in accordance with the principles of Good Clinical Practice (GCP).

In addition to Sponsorship approval, a favourable ethical opinion will be obtained from an appropriate NHS REC. Authorisation from the MHRA, and appropriate NHS R&D permission(s) will be obtained prior to commencement of the trial.

14.2 CONFIDENTIALITY

The CI and trial staff will comply with the requirements of the Data Protection Act 2018 and the General Data protection Regulation with regard to the collection, storage, processing and disclosure of personal information and will uphold the Directive's core principles.

The CI and trial staff will also adhere to the NHS Scotland Code of Practice on Protecting Participant Confidentiality or equivalent.

All trial records and data will be managed in a manner designed to maintain participant confidentiality. All records, electronic or paper, will be kept in a secure storage area with access limited to appropriate trial staff only. Computers used to collate data will have limited access measures via user names and passwords.

Personal clinical information will not be released without the written permission of the participant, except as necessary for monitoring and auditing by the Sponsor, its designee or regulatory authorities.

The CI and trial staff will not disclose or use for any purpose other than performance of the trial, any data, record, or other unpublished, confidential information disclosed to those individuals for the purpose of the trial. Prior written agreement from the Sponsor will be required for the disclosure of any said confidential information to other parties.

Access to collated participant data will be restricted to the CI and appropriate delegated trial staff.

Where data requires to be transferred, an appropriate Data Transfer Agreement will be put in place.

Published results will not contain any personal data that could allow identification of individual participants.

14.3 INSURANCE AND INDEMNITY

The University of Dundee and Tayside Health Board are Co-Sponsoring the trial.

Insurance. – The University of Dundee will obtain and hold Professional Negligence Clinical Trials Insurance cover for legal liabilities arising from the trial.

Tayside Health Board will maintain its membership of the Clinical Negligence and Other Risks Insurance Scheme ("CNORIS") which covers the legal liability of Tayside in relation to the trial.

Where the trial involves University of Dundee staff undertaking clinical research on NHS participants, such staff will hold honorary contracts with Tayside Health Board which means they will have cover under Tayside's membership of the CNORIS scheme.

Indemnity. The Co-Sponsors do not provide trial participants with indemnity in relation to participation in the Trial but have insurance for legal liability as described above.

Where other Scottish Health Boards are participating as trial sites, those other Scottish Health Boards will maintain membership of CNORIS to cover their liability in relation to their conduct of the trial.

Where other UK NHS organisations are participating as trial sites, those other UK NHS organisations will maintain membership of a scheme similar to CNORIS.

14.4 PROTOCOL AMENDMENTS

Amendments to protocol will be conducted in compliance with Sponsor SOP. The CI will seek Sponsor approval for any amendments to the Protocol or other approved trial documents. Amendments to the protocol or other trial documents will not be implemented without approval from the Sponsor and subsequent approval from the appropriate REC and/or MHRA, as appropriate, and NHS R&D Office(s).

14.5 PROTOCOL DEVIATIONS, BREACHES AND WAIVERS

Management of breaches to protocol or GCP will be conducted in compliance with Sponsor SOP

The CI will not implement any deviation from the protocol without agreement from the Sponsor, except where necessary to eliminate an immediate hazard to trial participants.

In the event that there is a deviation from the protocol, the nature of and reasons for the deviation will be recorded in the CRF/TMF and documented in the trial TASC Breach Log.

If a serious breach of the protocol or GCP is suspected, this will be reported to the Sponsor immediately using the TASC Breach Reporting Form and will be recorded in the CRF and documented in the trial TASC Breach Log.

If a breach necessitates a subsequent protocol amendment, this will be submitted to the Sponsor for approval and categorisation and then to the appropriate REC, MHRA and NHS R&D for review and approvals as appropriate.

It is Sponsor policy that waivers to the Protocol will not be approved.

14.6 TRIAL RECORD RETENTION

Archiving of trial documents will be as detailed in the archiving plan, carried out as specified Sponsor SOP. All trial documentation, electronic and paper, will be kept for 5 years. Case notes will be maintained in compliance with local NHS Policy on Retention of Medical Case notes.

14.7 END OF TRIAL

The end of trial at all Sites is defined as last database lock. The Sponsor and CI have the right at any time to terminate the trial for clinical or administrative reasons.

The end of the trial will be reported to the Sponsor, REC, MHRA and NHS R&D Office(s) within 90 days, or 15 days if the trial is terminated prematurely. The CI will ensure that any appropriate follow up is arranged for all participants.

A final clinical trial report will be submitted to the MHRA via EudraCT within 1 year of the end of the trial and will also be provided to the Sponsor and REC.

15. REPORTING, PUBLICATIONS AND DISSEMINATION OF RESULTS

15.1 AUTHORSHIP POLICY

The data arising from this trial resides with the trial team and ownership with the University of Dundee. On completion of the trial, the trial data will be analysed and tabulated, and a clinical trial final report will be prepared.

15.2 PUBLICATION

Details of the trial and clinical trial final report will be published on the EudraCT database, the latter no later than 12 months after the end of trial and will be available to the public via the EU Clinical Trial Register. The report will be made available to the Funder. The report can be used for publication and presentation at scientific meetings. Trial investigators have the right to publish orally or in writing the results of the trial. The criteria for authorship will follow the criteria of The International Committee of Medical Journal Editors as described in the TASC Publication Policy.

The clinical trial report will be used for publication and presentation at scientific meetings. Trial Investigators have the right to publish orally or in writing the results of the trial.

Publications will be reviewed according to the agreed contractual terms but will not restrict the general rights outlined above for the Investigators to publish the results of the trial.

Summaries of results will also be made available to Investigators for dissemination within their clinical areas (where appropriate and according to their discretion).

15.3 PEER REVIEW

This trial has been funded by Gilead who have reviewed the grant application, and the protocol has been reviewed and approved by the Sponsor Committee responsible for this.

Resulting publications will be reviewed by the referees of the journal to which the paper (and its protocol) will be submitted.

16. REFERENCES

Barker AF, Couch L, Fiel SB, Gotfried MH, Ilowite J, Meyer KC, ODonnell A, Sahn SA, Smith LJ, Stewart JO, Abuan T, Tully H, Van Dalfsen J, Wells CD, Quan J. Tobramycin solution for inhalation reduces sputum Pseudomonas aeruginosa density in bronchiectasis. *Am. J. Respir. Crit. Care Med.* 2000; 162: 481–485.

Barker AF, O'Donnell AE, Flume P, Thompson PJ, Ruzi JD, De Gracia J, Boersma WG, De Soyza A, Shao L, Zhang J, Haas L, Lewis SA, Leitzinger S, Montgomery AB, McKevitt MT, Gossage D, Quittner AL, O'Riordan TG. Aztreonam for inhalation solution in patients with non-cystic fibrosis bronchiectasis (AIR-BX1 and AIR-BX2): Two randomised double-blind, placebo-controlled phase 3 trials. *Lancet Respir. Med.* 2014; 2: 738–749

Chalmers JD, Moffitt KL, Suarez-Cuartin G, Sibila O, Finch S, Furrie E, et al. Neutrophil Elastase Activity Is Associated with Exacerbations and Lung Function Decline in Bronchiectasis. American journal of respiratory and critical care medicine. 2017;195(10):1384-93.

Finch S, McDonnell MJ, Abo-Leyah H, Aliberti S, Chalmers JD. A Comprehensive Analysis of the Impact of Pseudomonas aeruginosa Colonization on Prognosis in Adult Bronchiectasis. Annals of the American Thoracic Society. 2015;12(11):1602-11

Chalmers JD, Smith MP, McHugh BJ, Doherty C, Govan JR, Hill AT. Short- and long-term antibiotic treatment reduces airway and systemic inflammation in non-cystic fibrosis bronchiectasis. *Am. J. Respir. Crit. Care Med.* 2012; 186: 657–665

Haworth CS, Foweraker JE, Wilkinson P, Kenyon RF, Bilton D. Inhaled colistin in patients with bronchiectasis and chronic pseudomonas aeruginosa infection. *Am. J. Respir. Crit. Care Med.* 2014; 189: 975–982.

Haworth CS, Bilton D, Chalmers JD, Davis AM, Froehlich J, Gonda I, Thompson B, Wanner A, O'Donnell AE. Inhaled liposomal ciprofloxacin in patients with non-cystic fibrosis bronchiectasis and chronic lung infection with Pseudomonas aeruginosa (ORBIT-3 and ORBIT-4): two phase 3, randomised controlled trials. *Lancet Respir Med*. 2019 Jan 15. pii: S2213-2600(18)30427-2. doi: 10.1016/S2213-2600(18)30427-2

Kidd TJ, Canton R, Ekkelenkamp M, Johansen HK, Gilligan P, LiPuma JJ, Bell SC, Elborn JS, FlumePA, VanDevanter DR, Waters VJ; Antimicrobial Resistance in Cystic Fibrosis International Working Group. Defining antimicrobial resistance in cystic fibrosis.*J Cyst Fibros.* 2018 Nov;17(6):696-704. doi: 10.1016/j.jcf.2018.08.014. Epub 2018 Sep 25. Review.

Murray MP, Govan JRW, Doherty CJ, Simpson AJ, Wilkinson TS, Chalmers JD, Greening AP, Haslett C, Hill AT. A randomized controlled trial of nebulized gentamicin in non-cystic fibrosis bronchiectasis. *Am. J. Respir. Crit. Care Med.* 2011; 183: 491–499

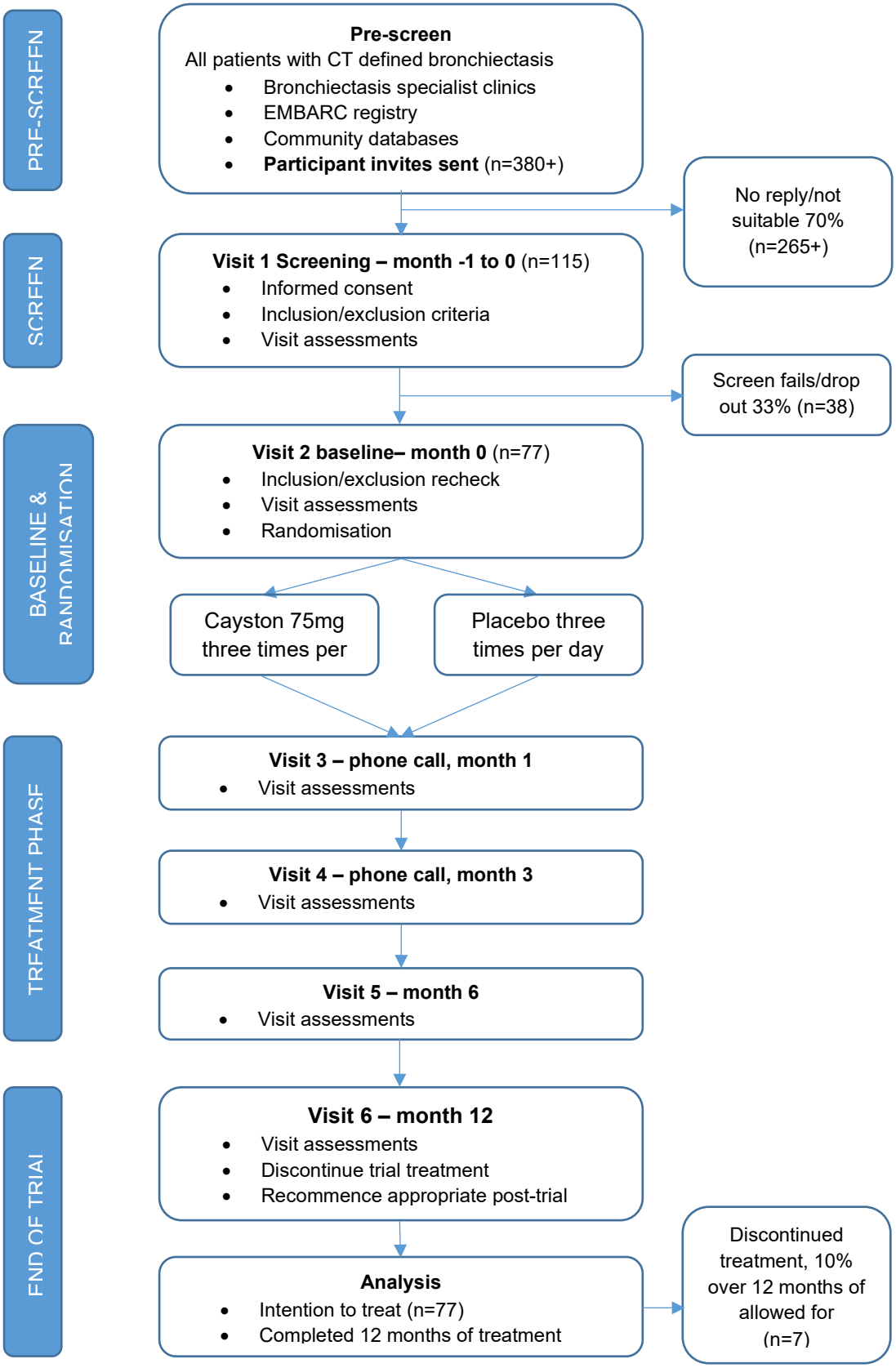
Nadig TR, Flume PA. Aerosolized antibiotics for patients with bronchiectasis. *Am J Respir Crit Care Med* 2016; 193(7):808-10.

Oermann CM, McCoy KS, Retsch-Bogart GZ, Gibson RL, McKevitt M, Montgomery AB. Pseudomonas aeruginosa antibiotic susceptibility during long-term use of aztreonam for inhalationsolution (AZLI). *J Antimicrob Chemother*. 2011 Oct;66(10):2398-404. doi: 10.1093/jac/dkr303. Epub 2011 Jul 22.

Somayaji R, Parkins MD, Shah A, Martiniano SL, Tunney MM, Kahle JS, Waters VJ, Elborn JS, Bell SC, Flume PA, VanDevanter DR; Antimicrobial susceptibility testing (AST) and associated clinical outcomes in individuals with cystic fibrosis: A systematic review. Antimicrobial Resistance in Cystic Fibrosis InternationalWorking Group. *J Cyst Fibros.* 2019 Jan 30. pii: S1569-1993(19)30014-1. doi: 10.1016/j.jcf.2019.01.008. [Epub ahead of print]

Taylor SL, Rogers GB, Chen AC, Burr LD, McGuckin MA, Serisier DJ. Matrix metalloproteinases vary with airway microbiota composition and lung function in non-cystic fibrosis bronchiectasis. Ann Am Thorac Soc. 2015 May;12(5):701-7. doi: 10.1513/AnnalsATS.201411-513OC

APPENDIX 1: TRIAL FLOWCHART & CONSORT DIAGRAM



APPENDIX 2: TRIAL MATRIX

Type of visit	Screening V1	Baseline and randomization V2	Follow-up phone call V3	Follow-up phone call V4	Follow-Up Assessments V5	Final visit Assessments V6	Unscheduled visit Assessments
Timeline	Month -1 to 0	Up to 35 days after screening	Month 1 (4 weeks) (+/- 2 Weeks)@	Month 3 (12 weeks) (+/- 2 Weeks)@	Month 6 (24 weeks) (+/-2 Weeks)@	Month 12 (48 weeks) (+/- 2 weeks)@	As Required
Informed Consent	X						
Inclusion/Exclusion Criteria Check	X	X					
Medical History	X						
Record Concomitant Medications	X	X	X	X	X	X	X
Physical Examination	X						X
Height	X						
Weight	X						
Check Vital Signs^	X	X			X	X	X
ECG	X						
Full blood count	X						
Urea and electrolytes	X						
Liver function tests	X						
Sputum samples for C+S and antibiotic resistance testing#	X				X	X	X

Type of visit	Screening V1	Baseline and randomization V2	Follow-up phone call V3	Follow-up phone call V4	Follow-Up Assessments V5	Final visit Assessments V6	Unscheduled visit Assessments
Timeline	Month -1 to 0	Up to 35 days after screening	Month 1 (4 weeks) (+/- 2 Weeks)@	Month 3 (12 weeks) (+/- 2 Weeks)@	Month 6 (24 weeks) (+/-2 Weeks)@	Month 12 (48 weeks) (+/- 2 weeks)@	As Required
Research Blood Sample *		X			X	X	X
Sputum sample for storage		X			X	X	X
Standard Spirometry	X	X			X	X	X
Quality of life bronchiectasis questionnaire&		X	X&	X&	X&	X	X
SGRQ		X	X	X	X	X	
BHQ		X	X	X	X	X	X
Exacerbation recording		X	X	X	X	X	X
Viral nasal swab		X				X	X
Supervised first dose of trial medication		X					
Safety Spirometry Post trial medication		X					
Bronchiectasis severity index and components e.g., MRC dyspnoea score	X						
Pregnancy Testing If Applicable	X	X			X	X	X
Record Adverse Events		X	X	X	X	X	X

Type of visit	Screening V1	Baseline and randomization V2	Follow-up phone call V3	Follow-up phone call V4	Follow-Up Assessments V5	Final visit Assessments V6	Unscheduled visit Assessments
Timeline	Month -1 to 0	Up to 35 days after screening	Month 1 (4 weeks) (+/- 2 Weeks)@	Month 3 (12 weeks) (+/- 2 Weeks)@	Month 6 (24 weeks) (+/-2 Weeks)@	Month 12 (48 weeks) (+/- 2 weeks)@	As Required
Randomisation		X					
Dispense Trial Drugs**		X[a]	X[a]	X[a]	X[a]		
Courier drugs to participant			X	X			
Drug Return And Compliance Check			X	X	X	X	

Under exceptional circumstances (*which may include: in the event of another lockdown, it is felt unsafe for the patient to come in as they should be self-isolating, patient declines visit due to COVID risk*) visits 5, 6 and unscheduled visits can be performed remotely e.g over the telephone.

^ Vital Signs: Blood Pressure, pulse, temperature, oxygen saturation

Where rapid PCR using the Biofire PCR system is available (e.g., Tayside site) this may be used to confirm the presence of protocol defined Gram negative pathogens to allow combined screening and randomization in the same day (the Biofire system takes approximately 1 hour to obtain a result). If there is sufficient sputum an aliquot will still be send for culture and sensitivity testing, but the PCR is sufficient to confirm eligibility.

@trial visits will be delayed after an unscheduled visit; they will be rescheduled for 7 days following the participant's recovery from the exacerbation

*Research blood samples will be stored and analysed at the end of the trial

a Trial drugs may be dispensed on a monthly basis depending on the preference of the participant.

**The 28-day off trial drug cycle maybe amended by the appropriate number of days to get a patient back on track for the 28-day treatment cycle. This may occur in situations such as participant going on a planned holiday. Trial teams to discuss with CI in such situations.

& Quality of life bronchiectasis questionnaire will be completed each month.

APPENDIX 3 Protocol Amendments

Amendment No.	Protocol Version No.	Author(s) of changes	Brief Details of Changes made
1	2	S. Inglis	Prebronchodilator spirometry removed from visit2. Screening period extended from 28 to 35 days.
2	3	S. Inglis	Update SmPC to newer version (5/6/18)
Response to REC	4	S. Inglis	Responses to REC review: • Clarification of process for antibiotic prescription during exacerbation (Section 4.4.1) • Info about research blood assessments (Section 4.4.6) • Clarification of restricted medications (Section 8.2.2)
Response to MHRA	5	S. Inglis	Responses to MHRA review: • Clarification of risk assessment around likelihood of antibiotic resistance development (Section 13.4.2, 13.4.3, 13.4.4, 4.1, 3) • Additional sputum culture and antibiotic resistance test at 6 months. • Further info about hypersensitivity to IMP (summary and section 5.3) • Contraception (Section 5.4) • Pregnancy testing (serum for eligibility) (Section 4.5.5) • Allergic reactions management (Section 5.3 and 7.1.7) • Info about discontinuation (Section 7.1.7)
AM04	6	S. Inglis	• We would like to ask participants to complete the QoL – B questionnaire every month during the study to ensure that we capture QoL after on- and off- treatment

			periods. Matrix updated to show this. Section 3 table 2 updated to show this. Text added section 4.4.4. • Nurses requested that they are able to phone participants to remind them to start their month's medication. Text added section 4.1 • Target sample size changed from 100 to 102 so that figures in each group add up to total (see section 4.1) • Clarification that results from blood/sputum samples taken within 5 days of the visit will be valid for the study to prevent the requirement for further blood/sputum samples to be taken. (Section 4.4). • Indication that time to first exacerbation outcome should be a primary outcome (primary efficacy objective), not secondary (section 3). Previous protocol versions alluded to this but time to first exacerbation was listed as a secondary outcome. Primary safety objective remains as in previous protocol versions. • Protocol V6 01/05/2019 submitted to sponsor on 3/5/2019. Shortly thereafter, CI noticed a typographical error in section 13.4.2 where amikacin should be replaced with aztreonam. This change was made and V6 15/5/2019 was submitted to sponsor on 15/5/2019.
AM07	7	C Clarke	• New participant pathway during COVID-19 outbreak. • This is a safety measure to safeguard participants. This measure was strongly recommended by the infectious disease-respiratory medicine working group on COVID-19 within NHS Tayside.
AM08	8	C Clarke	• Antibiotic resistance testing should be done on the screening sample that grows the bug. Use of the word 'baseline' was causing some confusion as 'screening' relates to visit 1 and 'baseline' relates to visit 2 (randomisation visit). • Shortening of 28-day IMP off cycle to allow for full 28-day treatment cycles (*for participant convenience in the event of planned holidays for example*). Recent breaches regarding this, so amendment to protocol as advised by Sponsor.

			• To clarify, participants who did not have a prescribed bronchodilator will be prescribed one at site (treatment cost). • CRFs for screen fails were not being completed as fully as possible as recording assessments stopped at the point of screen fail. To clarify, all screening assessments should be carried out for screen fail participants (*except for taking blood or inducing sputum*) and CRF sheets for this visit must be otherwise completed in full.
AM09	9	M Band/J Chalmers	• Removal of 75mg Cayston/placebo two times per day arms • Reduction in total number of participants to be recruited. • Removal of sputum induction • Change follow up visits at 1 and 3 months from face-to-face to telephone contact. • Addition of the use of PCR, where available, to confirm the presence of protocol defined Gram negative pathogens. This may allow combined screening and randomisation in the same day • Change of data management system from OpenClinica to Castor. • Removal of Appendix 4 describing new participant pathway during COVID19 pandemic.
AM15	10	F McLaren-Neil	• Secondary outcome measure amended to match changes in trial design AM09 • Correction of typos and grammatical errors
AM16	11	F McLaren-Neil	• Clarification of inclusion criteria *Pseudomonas aeruginosa or other Gram-negative respiratory pathogen detected in sputum or bronchoalveolar lavage on at least 1 occasion in the previous 12 months;* A potential participant who has no sputum sample in the previous 12 months can be invited for screening and if a positive sample is obtained at the screening visit meets this eligibility criteria.