



FULL/LONG TITLE OF THE TRIAL

A randomised, open-label, multifactorial, multicentre, platform trial using a range of repurposed anti-inflammatory treatments to improve outcomes in patients with bronchiectasis within the EMBARC clinical research network.

SHORT TRIAL TITLE / ACRONYM

AIR-NET- Testing anti-inflammatories for the treatment of bronchiectasis

PROTOCOL VERSION NUMBER AND DATE

AIR-NET Protocol V4 21-01-2025

RESEARCH REFERENCE NUMBERS

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SIGNATURE PAGE

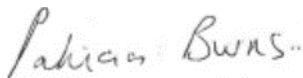
The undersigned confirm that the following protocol has been agreed and accepted and that the Chief Investigator agrees to conduct the trial in compliance with the approved protocol and will adhere to the principles outlined in the Medicines for Human Use (Clinical Trials) Regulations 2004 (SI 2004/1031), amended regulations (SI 2006/1928) and any subsequent amendments of the clinical trial regulations, Good Clinical Practice (GCP) guidelines, the Sponsor's (and any other relevant) Standard Operating Procedures, and other regulatory requirements as amended.

I agree to ensure that the confidential information contained in this document will not be used for any other purpose other than the evaluation or conduct of the clinical investigation without the prior written consent of the Sponsor.

I also confirm that I will make the findings of the trial publicly available through publication or other dissemination tools without any unnecessary delay and that an honest accurate and transparent account of the trial will be given; and that any discrepancies and serious breaches of GCP from the trial as planned in this protocol will be explained.

For and on behalf of the Trial Sponsor:

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29.01.2025

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27/01/2025

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Statistician

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I. LIST OF ABBREVIATIONS

AE	Adverse Event
AR	Adverse Reaction
BEST	Bronchiectasis exacerbation and symptom tool
BIM	Bronchiectasis Impact Measure
bPIS	Brief Participant Information Sheet
CI	Chief Investigator
CNORIS	Clinical Negligence and Other Risks Insurance Scheme
COPD	Chronic Obstructive Pulmonary Disease
DMC	Data Monitoring Committee
ECG	Electrocardiogram
eCRF	Electronic Case Report Form
EDC	Electronic Data Capture
EMBARC	European Multicentre Bronchiectasis Audit and Research Collaboration
FDA	US Food & Drug Administration
GCP	Good Clinical Practice
GDPR	General Data Protection Regulation (EU) 2016/679
GSDMD	Gasdermin D
IMP	Investigational Medicinal Product
ISF	Investigator Site File (This forms part of the TMF)
ISRCTN	International Standard Randomised Controlled Trials Number
FLPI	Full-field Laser Perfusion Imaging
LPLV	Last Participant Last Visit
MA	Marketing Authorisation
MAMS	Multi-arm, multistage trials
MHRA	Medicines and Healthcare products Regulatory Agency
MMP	Matrix-metalloproteinases
NE	Neutrophil elastase
NEATstik	Neutrophil Elastase Airways Test
NETs	Neutrophil extracellular traps
NHS R&D	National Health Service Research & Development
NSP	Neutrophil serine proteases
PI	Principal Investigator
PIS	Participant Information Sheet
PWV	Pulse Wave Velocity
RSI	Reference Safety Information
QoL-B	Quality of Life Bronchiectasis
REC	Research Ethics Committee
SAE	Serious Adverse Event
SDV	Source Data Verification
SmPC	Summary of Product Characteristics
SUSAR	Suspected Unexpected Serious Adverse Reaction
TASC	Tayside Medical Science Centre
TCTU	Tayside Clinical Trials Unit
TRuST	Tayside Randomisation System
TMF	Trial Master File
TMG	Trial Management Group
TSC	Trial Steering Committee
UoD	University of Dundee
WOCBP	Women of childbearing potential

II. TRIAL SUMMARY

Trial Title	A randomised, open-label, multifactorial, multicentre, platform trial using a range of repurposed anti-inflammatory treatment to improve outcomes in patients with bronchiectasis within the EMBARC clinical research network.	
Short title	AIR-NET- Testing anti-inflammatories for the treatment of bronchiectasis	
Clinical Phase	II	
Trial Design	Randomised, open-label, multifactorial, multicentre, platform trial	
Trial Participants	Bronchiectasis patients with active neutrophilic inflammation	
Planned Sample Size	Sample size for each arm is given below.	
Treatment duration	28 days	
Follow up duration	56 days	
	Objectives	Outcome Measures
Primary	To evaluate the effect of a range of interventions compared to usual care on the activity of Neutrophil elastase in sputum	Sputum neutrophil elastase activity at day 28
Control arm	Arm 1: Usual care Participants will be contemporaneously randomised to the control arm until trial end	
Open treatment arms	Arm 2: n=42, disulfiram - two 200mg oral tablets once daily Arm 3: n=42, dipyridamole – one 200mg oral prolonged/modified release capsule twice daily Arm 4: n=42, doxycycline – one 100mg oral capsule once daily Additional treatment arms will be added as identified	
Closed treatment arms	Nil, initial trial protocol, no treatment arms have been closed.	
Sub-study		
Trial Participants	Participants recruited to the main trial at the Tayside site will be enrolled in the sub-study	
	Objectives	Outcome measures
Exploratory	To evaluate the effect of a range of interventions compared to usual care on the cardiovascular system	Pulse wave velocity Full-field Laser Perfusion Imaging

III. FUNDING AND SUPPORT IN KIND

FUNDER(S)	FINANCIAL AND NON FINANCIAL SUPPORT GIVEN
LifeArc	Funding

IV. ROLE OF TRIAL SPONSOR AND FUNDER

The roles and responsibilities of the Sponsor and Funder will be detailed in the Clinical Research Agreement.

V. ROLES AND RESPONSIBILITIES OF TRIAL MANAGEMENT COMMITTEES/GROUPS & INDIVIDUALS

The trial will be coordinated by a Trial Management Group (TMG), consisting of the grant holders, including the CI, collaborators, statistician, research assistant, trial manager and research nurse where appropriate. TMG membership details will be held in the Trial Master File (TMF). The TMG will meet regularly to ensure all practical details of the trial are progressing well and working well and everyone within the trial understands them. Minutes of the TMG meetings will be maintained in the TMF.

The trial will have an independent Trial Steering Committee (TSC). The role of the TSC will be detailed in a TSC charter and will include oversight of the conduct of the trial including decisions around stopping or adding trial arms. TSC terms of reference are detailed in the TSC Charter and held in the TMF. Minutes of the TSC will be maintained in the TMF.

A Data Monitoring Committee (DMC) will be established to oversee the safety of trial participants and will be independent of the Sponsor. The DMC will be unblinded to allocation. The DMC will meet prior to participant recruitment to decide on the frequency of DMC meetings, timings will be documented in the DMC charter. DMC terms of reference are detailed in the DMC Charter and held in the TMF. Minutes of the DMC will be maintained in the TMF.

The Chief Investigator (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 Log will be prepared for each trial site, detailing the duties of each member of staff working on the trial.

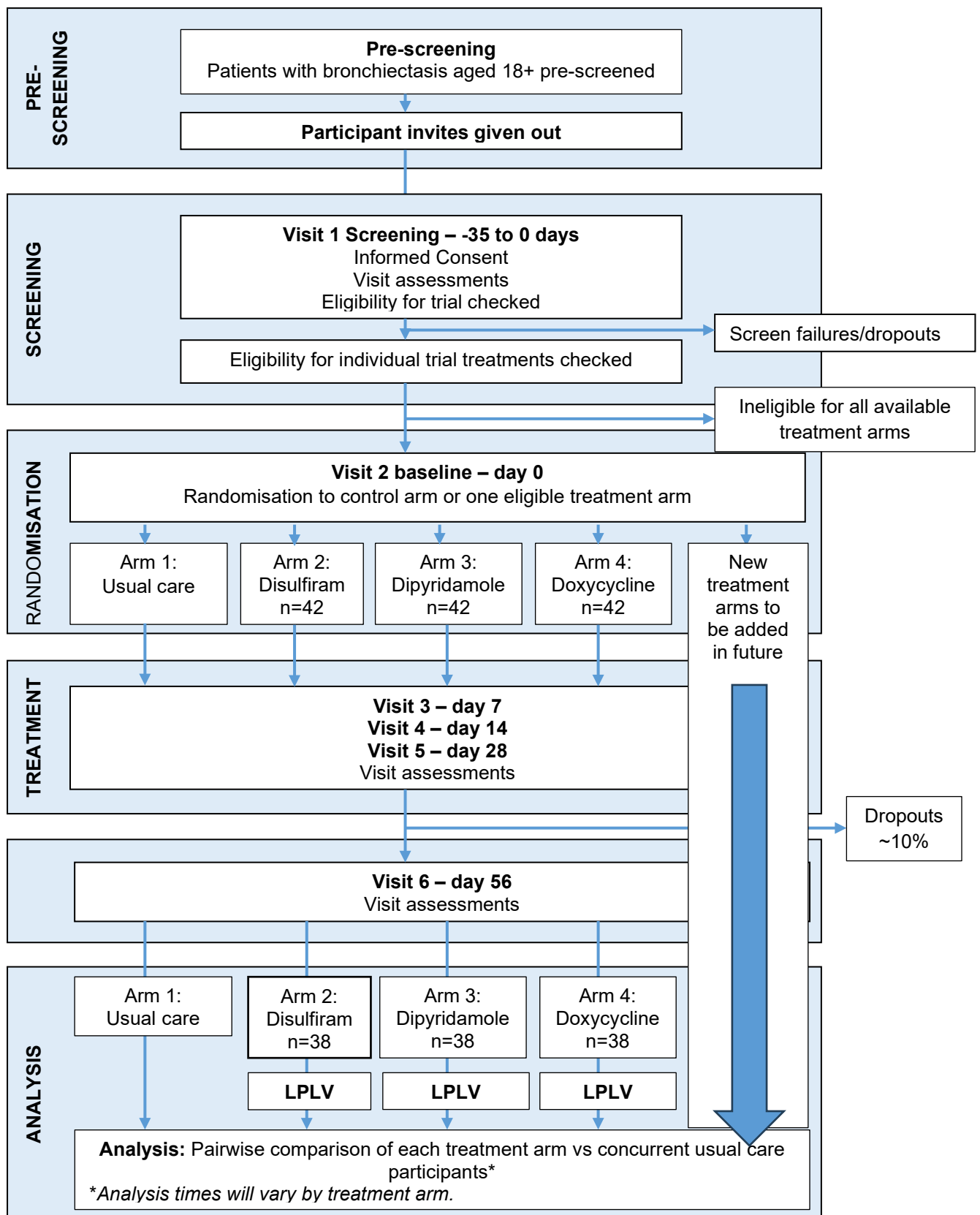
VI. PROTOCOL CONTRIBUTORS

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Senior Research Statistician: Jamie Stobo, review
Lead Scientist: Dr Merete Long, review
Clinical Trial Pharmacist: Shona Carson, review
TCTU Database Manager: Hasithi Umagiliya Bandara, review

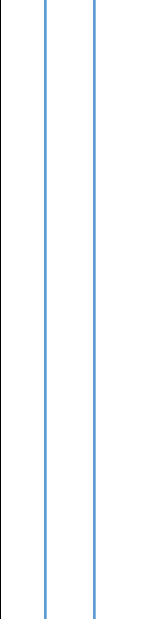
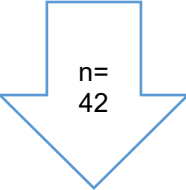
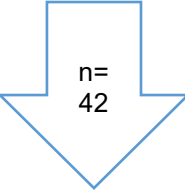
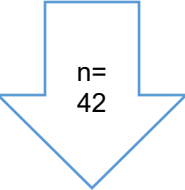
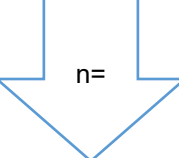
VII. KEY WORDS:

Bronchiectasis; anti-inflammatory; platform trial

VIII. PARTICIPANT FLOW CHART



IX. TRIAL TIMELINES FLOW CHART

Open/close to recruitment					New arms to be added in future	
Sept 2024	Arm 1: Usual Care 	Arm 2: Disulfiram  n= 42 OPEN	Arm 3: Dipyridamole  n= 42 OPEN	Arm 4: Doxycycline  n= 42 OPEN		
<div style="border: 1px solid black; padding: 5px; width: fit-content;"> Dates arms closed/new arms opened will be added </div>						
	OPEN					Arm: 6 XXX  n= N/A

1. BACKGROUND

Non-cystic bronchiectasis (hereafter referred to as bronchiectasis) is a chronic inflammatory disease with permanent bronchi dilatation, characterised by disabling productive cough, shortness of breath, and occasional haemoptysis (Barker 2002). Bronchiectasis is a heterogeneous condition that can either be a suppurative pulmonary disease on its own or overlap with other pulmonary diseases including asthma and chronic obstructive pulmonary disease (Chalmers et al. 2018). Data from the European Multicentre Bronchiectasis Audit and Research Collaboration (EMBARC) registry shows that approximately half of the patients with bronchiectasis experience two or more exacerbations a year, a major adverse event within the natural history of this disease. Patients who frequently exacerbate have double the mortality rate than patients who do not (Chalmers et al. 2014). Therefore, reducing exacerbations has now been an area of focus for most bronchiectasis research involving therapeutics.

There are few novel therapies in development for the treatment of bronchiectasis due to a lack of validated targets, and due to a lack of therapies with proof of concept to support further clinical development. The dominant mechanism of disease in bronchiectasis is chronic neutrophilic inflammation, which is near universal in patients with this condition, and which has been linked to disease severity, future exacerbations, and lung function decline. Excessive neutrophilic inflammation through the formation of neutrophil extracellular traps (NETs) is therefore a key therapeutic target in bronchiectasis (Chalmers et al. 2017). Dipeptidyl peptidase-1 inhibitors, which directly target neutrophilic inflammation, were recently shown to prolong time to first exacerbation in a phase 2 trial in bronchiectasis (Chalmers et al. 2020). Exacerbation reduction was predicted by a reduction in neutrophil elastase (NE) activity in sputum (Chalmers et al. 2020). NE has been shown to be a valid biomarker across multiple studies in bronchiectasis (Chalmers et al. 2017).

NETs and neutrophilic inflammation are therefore key therapeutic targets and validated biomarkers, which predict clinical response, with which to test the effectiveness of new therapies. Drugs which reduce neutrophil recruitment to the airway such as CXCR2 antagonists and leukotriene B4 antagonists might increase infections (Chapman et al. 2009; Döring et al. 2013; De Soyza et al. 2015). Therefore, anti-inflammatory treatment needs to modulate, rather than “switch off” inflammation in the airways. The cellular pathways leading to NET formation in neutrophils are now well defined and there are many potential repurposed therapies which could target this mechanism.(Döring et al. 2013; Barnes 2015).

There are no good animal models of bronchiectasis and consequently it is hard to identify candidate therapies with strong pre-clinical evidence. This means that currently, there is not one outstanding candidate for a repurposed anti-inflammatory therapy in bronchiectasis. Therefore, an approach is needed that will allow the screening of multiple drugs to identify the most effective and well-tolerated candidate for further clinical development in large scale trials.

2. RATIONALE

Understanding of the causes and consequences of bronchiectasis exacerbations has been improved recently through the latest clinical and translational research. The causes of bronchiectasis exacerbations can be broadly attributed to bacterial infection, viral infection, environmental effects, and host inflammatory response (Gao et al. 2024).

Chronic neutrophilic inflammation is a dominant part of the host inflammatory response in bronchiectasis, and has been associated with disease severity, further exacerbations, and a decline in lung function. Neutrophilic inflammation occurs partly from the accumulation of excess activity of neutrophil serine proteases (NSPs), one of which is NE. NSPs are released by neutrophils during degranulation and NETs release (NETosis) (Keir and Chalmers 2022). Attenuating neutrophilic inflammation was shown to reduce exacerbation frequency and prolong time to first exacerbation in the WILLOW trial, a phase 2 trial involving a novel dipeptidyl peptidase-1 inhibitor, Brensocatib (Chalmers et al. 2020). The WILLOW trial also demonstrated that sputum NE reduction is associated with reduction in bronchiectasis exacerbations and is a valid biomarker for other similar bronchiectasis trials.

This has promoted increasing interest around attenuating host inflammatory responses to reduce bronchiectasis exacerbations, but more research is needed around the exact mechanisms, as well as to explore wider implications.

There are numerous existing therapeutics that have shown to have secondary anti-inflammatory effects beyond their original indication (Rakocevic et al. 2023, Patel et al. 2023). Instead of repurposing and testing these therapeutics one at a time, it is proposed to establish a platform trial to enable rapid testing of multiple therapeutics within the population of bronchiectasis patients.

These repurposed therapeutics have established real-world evidence based on their safety profile, side effects, pharmacokinetics and pharmacodynamics, drug–drug interactions, and experience in different patient populations. This enables us to conduct trials more safely and cost-effectively.

In other fields such as cancer research, the idea of a randomized screening design is widely accepted (Rubinstein, 2005). In such designs, a potential treatment is tested against a control (often standard care, or an active comparator) using relaxed type I and II error rates to reduce the required sample size. This provides a pragmatic approach to quickly obtain a (non-definitive) signal of efficacy, to be confirmed in a larger phase III study, while screening out sub-optimal treatments at an early stage.

Adaptive platform trials, such as multi-arm, multistage trials (MAMS), offer the potential to expedite the testing of new therapies with reduced cost compared to conventional placebo controlled, single arm, parallel group, randomised controlled trials. Utilising repurposed compounds with established safety records in clinical practice instils greater confidence in participants and investigators, when compared to new compounds with limited long-term data or experience. MAMS have gained widespread use in the cancer field and have become more prevalent since the COVID-19 pandemic. The RECOVERY trial serves a notable example for a MAMS, using repurposed compounds such as dexamethasone, colchicine, and azithromycin among their candidates. Ultimately, the RECOVERY trial yielded the world's first effective agent against COVID-19 (Horby et al. 2020).

AIR-NET is a multi-arm, adaptive platform trial designed to test the anti-inflammatory effects of multiple repurposed compounds in patients with bronchiectasis. Participants will be randomized to one of multiple potential interventions or usual care and treated for 28-days. The effect of treatment on sputum NE levels will be the primary efficacy outcome, and safety will be determined. The agents with the greatest evidence of target engagement and safety will be prioritised for further development. Embedded within the platform will be a

programme of standardisation, capacity building and translational research that will enhance the UK's capacity to conduct early phase bronchiectasis trials.

Participants will be randomized to one of the open treatment arms they have been deemed eligible for, or usual care. Treatment will be administered for 28 days, a period which should be sufficient to demonstrate robust anti-inflammatory effects based on previous studies (Chalmers et al. 2020). All compounds are not currently used in the treatment of bronchiectasis as long-term therapies but have in vitro or in vivo evidence that they target neutrophilic inflammation and are widely available generic drugs which could be rapidly repurposed if shown to be effective. Any repurposed compounds demonstrating an efficacy signal against usual care will then be considered for further large-scale trials. Compounds that do not demonstrate sufficient efficacy or have safety issues will not be studied further (Chalmers et al. 2020). The Trial Steering Committee will evaluate the trial data to determine which compounds should be prioritised for further trials, and will review applications/proposals to add new experimental arms to the trial.

A sub-study will also be included as an option for those participants enrolled in Tayside. Neutrophilic inflammation is implicated in increased cardiovascular risk in patients with bronchiectasis and our sub-study will constitute a pilot study to determine if the interventions have an effect on biomarkers of cardiovascular risk.

Rationale for open trial treatments:

Arm 2: Disulfiram

The carbamate derivative disulfiram, a US Food & Drug Administration (FDA)-approved drug for treating alcohol dependency, received increasing research attention during the COVID-19 pandemic as both a potential anti-viral agent (Jin et al. 2020) and NET inhibitor via inhibition of Gasdermin D (GSDMD), which has shown to be involved in the generation and release of NETs (Hu et al. 2020). GSDMD cleavage and activation is the result of inflammasome activation or alternatively NE activity, which in immune cells ultimately results in active IL-1 β release from the cell, which then triggers multiple downstream immune and inflammatory processes. This includes either pyroptosis in the case of myeloid cells or NETosis in the case of neutrophils (He et al. 2015). Work within the Chalmers group has shown that IL-1 β and also caspase-1 activity levels in sputum from bronchiectasis patients, indicative of classical NLRP3 inflammasome activation, are associated with severe disease, mucus purulence and mucociliary dysfunction (Perea et al. 2024). In a murine model of acute lung injury, disulfiram treatment reduced NETs and lung oedema and improved survival, and in a further investigation in SARS-CoV-2 infected hamsters similar results were observed with complementary downregulation of innate immune pathways (Adrover et al. 2022). Whilst in these models total IL-1 β in the lung tissue was not reduced, other authors have shown that disulfiram does not block IL-1 β generation or impact caspase-1 dependent canonical signalling but rather prevents IL-1 β release from the cell by blocking caspase 11-dependent GSDMD pore formation. Reduced levels of IL-1 β as well as other inflammatory cytokines have been demonstrated with disulfiram treatment in pre-clinical models of LPS-induced sepsis and in human macrophages, in addition to other inflammatory cytokines including TNF and IL-6 (Hu et al. 2020). However, in murine and human macrophage cell lines in vitro in one screening study disulfiram was identified as a NLRP3 inflammasome pathway inhibitor, and reduced ASC-speck formation implicated in classical inflammasome formation and activity (Bonnekoh et al. 2021). These processes may differ between patients and animal models utilised and may not yet be fully elucidated, however disulfiram consistently

demonstrated significant beneficial effects via GSDMD inhibition. Further, disulfiram treatment was also shown to reduce histone H3 processing in neutrophils after LPS stimulation, a process concomitant with NETosis (Adrover et al. 2022). GSDMD is involved in pore-formation in neutrophil granules intracellularly, which releases NE that then acts in a positive feedback loop to activate GSDMD and promote NETosis (Sollberger et al. 2018), and reduced spontaneous NETosis when neutrophils from septic patients were treated directly with disulfiram in vitro (Silva et al. 2021). In cancer cells including malignant T-cells, disulfiram treatment has been shown to induce apoptosis and inhibit proliferation (Chen et al. 2022), these mechanisms may be cell-type specific and there is so strong evidence that apoptosis is promoted in other non-cancer cell types. On the contrary, in addition to reducing NETosis, disulfiram can act on other immune cell types to reduce harmful methods of cell death including the explosive macrophage pyroptosis. Neutrophils are not the only player in the lungs in bronchiectasis and are influenced significantly by the inflammatory environment they encounter. By impacting the proinflammatory activity of multiple cell types neutrophilic inflammation may also be quiesced.

Based on existing clinical practice, where doses of >200mg are frequently used, as well as from safety data from clinical trials, there have been no major safety concerns identified with the use of 400mg disulfiram daily. While the SmPC states that the maximum Disulfiram dose is 200mg once daily, that is in a context of a treatment duration up to six months, for patients with alcohol dependency. In the British National Formulary and clinical practice the maximum dose goes up to 500mg once daily for the treatment of alcohol dependence over a similar period, under supervision. As the recruitment cohort are not individuals with alcohol dependency, as well as excluding individuals with any major cardiovascular disease/liver/psychiatric diseases, it is thought that there are no major safety concerns about using the dose of Disulfiram 400mg once daily dose for only 28 days.

Arm 3: Dipyridamole

Adenosine triphosphate and Adenosine diphosphate released extracellularly can be detected by G-coupled receptors to activate purinergic signalling cascades in cells including neutrophils, monocytes and platelets. Neutrophils have been shown to be major P2X7R expressors, a key Adenosine triphosphate receptor, in a model of *Streptococcus pneumoniae* infection model (Karmakar et al. 2016). Downstream events after receptor engagement include NLRP3 inflammasome activation and IL-1 β production (Karmakar, et al. 2016) as well as prompting initiation of coagulation pathways promoting thrombus formation. The FDA-approved drug Dipyridamole indicated typically for thrombosis prevention, activates the A2aR adenosine receptor, and has been shown to reduce neutrophil ROS formation and NETosis (Ali, et al. 2019), whilst also having beneficial effects to reduce platelet and also endothelial activation (Chakrabarti, et al. 2005). Utilising murine models, the action of dipyridamole in neutrophils was shown to be cAMP and PKA dependent, acting to inhibit adenosine reuptake into the cell and also to stabilise cAMP by inhibiting phosphodiesterase activity which degrades cAMP to potentiate downstream effects (Ali et al. 2019, Kanthi et al. 2020). Dipyridamole has demonstrated further activities potentially beneficial in bronchiectasis. Whilst many inflammatory pathways are overactive, in other chronic lung diseases, suppressed interferon responses have demonstrated contribution to viral infection susceptibility and associated with increased exacerbation frequency (Singanayagam, et al. 2019). Dipyridamole was shown to promote IFN signalling in murine leukocytes (Galabov and Mastikova 1982), an effect and potential mechanism warranting further investigation.

Arm 4: Doxycycline

The tetracycline derivative doxycycline is an FDA-approved antibiotic and inhibitor of extracellular matrix-degrading matrix-metalloproteinases (MMPs), with demonstrated activity against a range of MMPs shown to be increased in the airways in bronchiectasis. Amongst these are MMP-2 which has been associated with differential microbiome profiles in bronchiectasis in severe disease (Taylor et al. 2015) and MMP-9 linked with bronchiectasis progression (Garratt et al. 2015). In animal models of chronic lung disease, subsequent doxycycline treatment after disease development was able to reduce MMP-2 and -9 expression in the lungs as well as levels of the inflammatory cytokines TNF- α and IL-1 β (Hadzic et al. 2021) and neutrophil infiltration (Lee et al. n.d.). Furthermore whilst studies so far have been small and highlight the need for further investigation, a small exploratory study in patients with Chronic Obstructive Pulmonary Disease (COPD) receiving 3 months of doxycycline add-on therapy demonstrated marked reductions in MMP levels in addition to reduced markers of oxidative stress and increased antioxidant markers (Singh et al. 2019). Reduction in MMP activity with doxycycline occurs via its activity as a calcium and zinc chelator and ability to bind these ions, in addition to direct inhibition of latent pro-MMPs (Golub et al. 1998) making the measurement of MMPs a potentially attractive biomarker of drug efficacy, adherence and treatment response.

However, in a variety of immune cell types and *in vivo* models, doxycycline treatment has demonstrated anti-inflammatory effects which may be induced by a range of mechanisms, including modulation of NF κ B, Akt, MAPK and TACE activity, all of which are implicated in both acute infection and in immune dysfunction in chronic lung disease (Di Caprio et al. 2015). *In vitro*, doxycycline treatment of human peripheral blood mononuclear cells (PBMCs; consisting primarily of B and T lymphocytes) stimulated with bacterial endotoxin resulted in dose-dependent reduction in inflammatory cytokine production including IL-1 β and IL-6 (Krakauer and Buckley 2003) and monocytes stimulated with *Aggregatibacter* (Bostanci et al. 2011), and reductions at both the protein level and gene expression have been evidenced (Bode et al. 2014). Suppression of NF κ B and MAPK pathways may be responsible for this effect, and doxycycline treatment of LPS-stimulated microglial cells reduced cell activation, proinflammatory cytokine release, ROS and NO production. This study also showed that phosphorylation of p38 MAPK and also NF- κ B nuclear translocation was inhibited with doxycycline treatment (Santa-Cecilia et al. 2016). Reduction in NF- κ B activity has been demonstrated in multiple cell types after doxycycline. In cancer cells doxycycline treatment downregulated the effects of LPS stimulation of MMPs, as well as NF- κ B (Ogut et al. 2016) and this has also been demonstrated in murine macrophages (Zhang et al. 2017). In several cell lines the mechanism of action was demonstrated as reducing downstream phosphorylated I κ B- α and IKK-B levels (Alexander-Savino et al. 2016). NF κ B activation contributes to a myriad of inflammatory functions, namely increasing the production of proinflammatory proteins, in addition dysfunctional mechanisms including suppression of phagocytic processes and induction of apoptosis (Bai et al. 2019). Further upstream, as a potential mechanism of action for these effects, Doxycycline has been demonstrated to act on NF κ B activator protein kinase C (PKC) in an *in vitro* granuloma model (Webster, Toso, and Hegemann 1994). Specific effects of doxycycline in neutrophils however have been much lesser investigated. Whilst direct application of doxycycline to neutrophils *in vitro* has yielded mixed results, PKC and ROS activity, Akt and MAPK signalling as well as inflammasome activation have all been implicated in NETosis, and will

be critical to understand whether treatment either directly or indirectly results in a reduction in NETs in the airways of patients receiving doxycycline.

In addition to reducing NFkB and MAPK activation, doxycycline has been shown to reduce NLRP3 inflammasome activation in immune cells *in vitro* and *in vivo* (Zhang et al. 2017), with similar beneficial effects to those described for disulfiram.

2.1. Assessment and Management of Risk

This trial is categorised as: Type B = Somewhat higher than the risk of standard medical care

The trial treatments are all repurposed drugs already approved for use in conditions other than bronchiectasis.

Arm 1: Usual care

Arm 2: Disulfiram

Disulfiram is an alcohol deterrent compound licensed for use as an adjuvant in the treatment of drinking problems. The most common adverse reaction (AR) reported during treatment is alcohol reaction. Disulfiram–ethanol reactions often develop within 15 minutes after exposure to ethanol; symptoms usually peak within 30 minutes to 1 hour, and then gradually subside over the next few hours. Symptoms may be severe and life-threatening. Participants will be informed of the reaction risk and must agree to abstain from alcohol during treatment and for up to 14 days after discontinuation.

Arm 3: Dipyridamole

Dipyridamole is licensed for use in secondary prevention of ischaemic stroke and transient ischaemic attacks and as an adjunct to oral anti-coagulation for prophylaxis of thromboembolism associated with prosthetic heart valves. The most commonly reported adverse effects are headache, dizziness, diarrhoea, nausea (very common, $\geq 1/10$) and angina pectoris, vomiting, rash, myalgia (common, $\geq 1/100 < 1/10$).

Arm 4: Doxycycline

Doxycycline is licensed for use in the treatment of a variety of infections caused by susceptible strains of Gram-positive and Gram-negative bacteria and certain other micro-organisms. The most common adverse effects observed in patients receiving tetracyclines, including doxycycline, are hypersensitivity, headache, nausea/vomiting, photosensitivity reaction rash including maculopapular and erythematous rashes (common, $\geq 1/100 < 1/10$).

Management of risk is described in Appendix 1. However, this is the first use of these medications in this patient population, hence there is the *possibility* of additional unknown risks.

3. OBJECTIVES AND OUTCOME MEASURES/ENDPOINTS

3.1. Table of endpoints/outcomes

Primary Objective		
Objective	Outcome Measures	Timepoint(s)
To evaluate the effect of a range of interventions compared to usual care on the activity of NE in sputum	Activity of sputum NE	Day 0 and 28
Secondary Objectives		
Objective	Outcome Measures	Timepoint(s)
To evaluate the effect of a range of interventions compared to usual care on the activity of NE in sputum	Activity of sputum NE	Days 0, 7, 14 and 56
To evaluate the effect of a range of interventions compared to usual care on time to onset of first bronchiectasis exacerbation	Time to first pulmonary exacerbation (EMBARC definition)	Days 0 to 28
To evaluate the effect of a range of interventions compared to usual care on quality of life	Quality of life-bronchiectasis (QOL-B) respiratory symptom scale, Bronchiectasis Impact Measure (BIM) questionnaire	Days 0, 7, 14, 28 and 56
To evaluate the effect of a range of interventions compared to usual care on clinical benefits	Bronchiectasis exacerbation and symptom tool (BEST) diary	Days 0 to 28
To evaluate the effect of a range of interventions compared to usual care on walking distance	Distance covered during 6-minute walk	Day 0, 28
To evaluate the safety of a range of interventions compared with usual care	Frequency of adverse events (AEs) and serious adverse events (SAEs)	Days 0 to 56
To evaluate the effect of a range of interventions on peripheral blood neutrophil function	Phagocytosis of bacteria Reactive oxygen species generation Degranulation Ex-vivo formation of neutrophil extracellular traps Mass cytometry (endpoints may vary depending on the experimental arm)	Day 0 to 28

Tertiary Objectives		
Objective	Outcome Measures	Timepoint(s)
To evaluate the effect of a range of interventions compared to usual care on bronchiectasis exacerbations	Frequency of pulmonary exacerbations (EMBARC definition)	Days 0 to 28; Days 0 to 56
To develop and validate additional biomarkers in sputum that are linked with clinical outcomes of bronchiectasis	Measure the concentration of MMPs and NETs in sputum, as well as other biomarkers e.g proteomics, bacterial load, microbiome	Days 0, 7, 14,28 and 56
To develop and validate additional biomarkers in blood that are linked with clinical outcomes of bronchiectasis	Measure serum biomarkers of inflammation and redox status	Days 0, 7, 14, 28 and 56
To investigate the effect of a range of interventions on peripheral blood neutrophils	Peripheral blood neutrophil proteomics performed on isolated cells	Days 0 and 28
To explore and characterise additional changes in inflammatory response in both sputum and blood across the range of interventions and usual care	Label free liquid chromatography/mass spectrometry sputum identification and relative quantification of proteins	Day 0 and 28
To explore changes in peripheral blood transcriptomics between interventions and usual care	Peripheral blood transcriptomics	Day 0 and 28
To explore changes in epithelial cell transcriptomics between interventions and usual care	Nasal brushing transcriptomics	Day 0 and 28
Sub-study Objectives		
Objective	Outcome Measures	Timepoint(s)
To evaluate the effect of a range of interventions compared to usual care on endothelial dysfunction	Change in skin perfusion with iontophoresis of acetylcholine and sodium nitroprusside using full-field laser perfusion imaging	Days 0, 28 and 56
To evaluate the effect of a range of interventions compared to usual care on the cardiovascular system	Change in arterial stiffness index	Days 0, 28 and 56
	Change in pulse wave velocity	

4. TRIAL DESIGN

Platform trial.

Parallel group design. Randomised, open-label, multifactorial, multicentre.

The trial will open to recruitment with a control arm and the first 3 treatment arms. Individual treatment arms will be closed for randomisation when the target for number of participants has been reached, or earlier if advised by the DMC and closed to follow up after Last Participant Last Visit (LPLV) for that arm. As new possibilities for treatments are identified additional treatment arms will be opened. Any new treatment arms will be submitted as an amendment to the current protocol. Randomisation to the control arm will continue for the duration of the trial.

4.1. Sub-study design

Participants enrolled within the Tayside site may be given the option of participation in the sub-study to examine endothelial and cardiovascular outcomes. The results from the sub-study assessments will be combined with results from separate trials, conducted in people with bronchiectasis, not connected with this protocol, for analysis.

5. TRIAL SETTING

AIR-NET will be conducted in a clinical trial network of 10 centres with capabilities to measure clinical and laboratory endpoints using the same optimised protocols. We will work with sites during the set-up phase of the study to standardise laboratory protocols for blood and sputum processing. The centres will be based in secondary care settings within the NHS in the UK.

5.1. Sub-study

Participants enrolled at the Tayside site.

6. PARTICIPANT ELIGIBILITY CRITERIA

6.1. Inclusion criteria

- ≥ 18 years.
- Able to provide informed consent.
- Capable of complying with all trial procedures and of completing the trial, in the opinion of the investigator.
- Bronchiectasis, confirmed by computed tomography (CT), showing bronchiectasis in 1 or more lobes.
- Normally produces sputum daily.
- Able to provide a sputum sample at the screening visit or between screening and randomisation^a.
- Active neutrophilic inflammation at screening/baseline indicated by a positive NEATstik (Neutrophil Elastase Airways Test) result^b.

^aRepeat sputum samples may be provided during the screening period, if the sample taken during the screening visit is deemed to be of insufficient quality or quantity by the laboratory.

^bA positive NEATstik test is equivalent to a NE concentration of 8µg/ml in sputum using the Proaxis active NE immunoassay. If NEATstik is not available for screening, a frozen sputum sample will be shipped to the central laboratory in Dundee where the immunoassay will be performed and used to confirm eligibility.

6.2. Exclusion criteria

- Enrolled previously in the trial 3 times
- Respiratory infection or bronchiectasis exacerbation 4 weeks prior to screening and/or between screening and randomisation^c
- Antibiotic or corticosteroid 4 weeks prior to screening and/or between screening and randomisation^c
- Active allergic bronchopulmonary aspergillosis (defined by International Society for Human and Animal Mycology criteria) on steroids and/or anti-fungals
- Nontuberculous mycobacterial infection on antibiotic therapy
- Immunodeficiency on immunoglobulin replacement
- A primary diagnosis of COPD or asthma (a secondary diagnosis of COPD or asthma is permitted)
- Cystic fibrosis
- Active malignancy except non-melanoma skin cancer
- Currently taking brensocatib
- Use of any investigational drugs within five times of the elimination half-life after the last dose or within 30 days, whichever is longer. Current enrolment in non-interventional, observational studies will be allowed
- Currently pregnant or breast-feeding
- Women of childbearing age and not practicing an acceptable method of birth control, see section 8.11

^c In the event of a respiratory infection or bronchiectasis exacerbation during the screening period, the screening period may be extended, once only by up to 8 weeks, to ensure that randomisation occurs at least 4 weeks after the last dose of antibiotics is given.

6.3. Open treatment arm specific exclusion criteria

6.3.1. Arm 2: Disulfiram

- Currently on Disulfiram (patients should have a washout period of at least 30 days from last dose if they have previously received this medication)
- Hypersensitivity to Disulfiram
- Participant, or investigator objects to randomisation to Disulfiram
- Does not agree to cease consumption of alcohol during intervention and for 14 days following treatment discontinuation
- Chronic liver disease
- Alanine transaminase (ALT)>135 U/L at screening,
- Bilirubin >30 µmol/L at screening.
- Uncompensated cardiac failure
- Coronary artery disease (diagnosis of stable or unstable angina, previous myocardial infarction)
- Previous history of stroke or transient ischaemic attack
- Uncontrolled hypertension
- Hereditary problems of galactose intolerance, total lactase deficiency or glucose-galactose malabsorption

- Recent psychiatric exacerbation
- Any significant acute or chronic psychiatric condition, including severe personality disorder, psychotic disorder or suicide risk.
- Hypothyroidism
- Porphyria
- Diabetes Mellitus
- Epilepsy

6.3.2. Arm 3: Dipyridamole

- Currently on Dipyridamole (patients should have a washout period of at least 30 days from last dose if they have previously received this medication)
- Hypersensitivity to Dipyridamole
- Participant, or investigator, objects to randomisation to Dipyridamole
- Currently on dual antithrombotic therapy (aspirin or P2Y12 inhibitor plus anticoagulation)
- Current on direct oral anticoagulants (Dabigatran, Rivaroxaban, Edoxaban, Apixaban, Betrixaban or drugs in the same class) or long-term warfarin
- Any major trauma or haemorrhage including gastrointestinal bleeding, operation within the past 30 days
- Coagulation disorder
- Severe coronary artery disease (unstable angina, recent myocardial infarct in 30 days, decompensated/unstable severe left systolic dysfunction, uncontrolled heart failure)
- Myasthenia gravis

6.3.3. Arm 4: Doxycycline

- Currently on Doxycycline (patients should have a washout period of at least 30 days from last dose if they have previously received this medication)
- Hypersensitivity to Doxycycline
- Participant, or investigator, objects to randomisation to Doxycycline
- Myasthenia Gravis
- Systemic Lupus Erythematosus
- Chronic Liver Disease
- Porphyria
- Alcohol dependence
- Suspected Syphilis

7. TRIAL PROCEDURES

7.1. Recruitment

Anonymised information on participants who are not randomised will be collected for CONSORT reporting and includes:

- age,
- gender,
- ethnicity,
- the reason not eligible for trial participation, or if they are eligible but declined.

7.1.1. Participant identification

Identification of potentially eligible trial participants by the research or clinical teams 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 a member of the clinical care team or research team, if delegated by the clinical care team, and medical records checked to identify suitable participants. Potential participants 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. Contact at clinic or class will be by a member of the clinical care team or local clinician. Postage of invitation letters and bPIS will be carried out by the clinical care team or a member of the trial team on delegation from the clinical care team.
- 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 facilities as defined locally. Local Principal Investigator (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)
- From primary care via the Primary Care Networks. These participants will be sent an invitation letter and bPIS from the GP practice. GP practices will also be asked to display bPIS in their waiting rooms.

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. Where a patient does not contact the research team the research team may contact them once via telephone, email or post to determine their interest in the trial.

Should individuals express an interest in taking part in the trial, the PI or delegate will contact the individual and ask for 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. Participants will receive a full Participant Information Sheet (PIS).

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.

7.1.2. Screening

At the screening visit (Visit 1) the procedures as detailed in the Schedule of Procedures, Appendix 4, will be carried out.

Assessment of eligibility will be carried out by the PI or other medically qualified delegate. Eligibility will be assessed at Visit 2, randomisation/baseline once all results have been reviewed.

Details of all participants consented to the trial and screened for eligibility will be recorded on the Enrolment and Randomisation Log, this will detail if a participant fails screening or goes on to be randomised.

Where an ineligible participant's medical condition, including ineligibility due to a recent infection, or concomitant medications change sufficiently so that they are deemed potentially eligible for the trial they may be rescreened one further time. All screening procedures will be repeated, including informed consent, and eligibility checked.

7.1.3. 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 incidental finding (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.

7.1.4. Payment

Participants will receive £100 per visit attended to cover travel expenses and recompense for attending research visits after each visit or at the end of their trial duration.

7.2. Consent

The PI retains overall responsibility for the recording of informed consent of participants at their site. They will ensure that any person delegated the informed consent process is duly authorised, trained and competent to participate according to the ethically approved protocol, principles of GCP and Declaration of Helsinki.

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

For adults who lose capacity their previous wishes will remain legally binding, and this will remain valid unless the protocol changes significantly. If this occurs and further consent is required from a participant who has lost capacity, the participant's legal representative, or if not contactable, a professional legal representative will be asked for their consent.

In all cases the PI 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.

7.2.1. Additional consent provisions for collection and use of participant data and biological specimens in ancillary studies, if applicable

Consent from participants will be gained for:

- use of their data in future research
- use of their specimens in future research

- use of their specimens in genetic research
- contact by trial staff for further ethically approved future research (optional)

Anonymised trial data will be kept under the control of the CI for future research use within the University of Dundee (UoD) and with other research collaborators (commercial and non-commercial).

Blood and sputum will be stored for future research in labs at the UoD. Specimens may be used for ethically approved research held within Tayside and will be registered with NHS Tayside Biorepository. Access for future use of those specimens will be via the CI. An EDTA blood sample for future genetic research will be obtained and patients will give consent for future genetic studies.

Any data collected to the point of withdrawal will be retained for reasons of public interest in the area of public health (Article 9(2)(i) General Data Protection Regulation (EU) 2016/679 GDPR).

7.3. Randomisation scheme

Participants will be randomised equally to any open arms they are eligible for. Participants will be allocated to an arm using a minimisation algorithm, with factors for centre, *Pseudomonas aeruginosa* infection recorded in the last 2 years and long-term use of macrolides.

Randomisation will initially open with a control arm and 3 treatment arms. When a treatment arm has recruited to its target, randomisation to that arm will be closed. When a new treatment arm is added, randomisation to that arm will be opened for all participants meeting the eligibility criteria for that arm.

Participants who have completed 56 days of follow-up (end of trial) may be rescreened and re-randomized if they are willing to participate in the trial again. Participants will receive a new copy of the current PIS, will be asked to complete a new Informed Consent Form, be given a new, unique participant identification number and eligibility will be rechecked prior to randomisation. All trial activities will be carried out as per protocol. Re-randomisation will be performed in the same way as for initial randomisations. Patients can be randomised a maximum of three times (i.e. initial randomisation and two re-randomisations).

7.3.1. Method of implementing the randomisation/allocation sequence

After successful completion of screening the participant will be assessed for eligibility for each treatment arm. This will be documented in the participant's medical notes and electronic Case Report Form (eCRF).

Participants will be randomised by the PI or delegate as per eligibility to the open trial arms.

The PI or delegate will use a centrally controlled web-based GCP compliant randomisation system, Tayside Randomisation System (TRuST), run by the UK Clinical Research Collaboration registered Tayside Clinical Trials Unit (TCTU). TRuST is provided by the Health Informatics Centre, UoD. TCTU use a validated randomisation program and will securely backup both the randomisation seed and the randomisation allocation.

The randomisation allocation will be emailed to the person completing the randomisation and Clinical Trials Pharmacy.

Access to be able to randomise a participant will only be given after completion of appropriate training.

7.4. Blinding

Nil, open label.

7.5. Emergency Unblinding

Not applicable

7.6. Baseline data

Baseline data will be collected at day 1 as per Schedule of Procedures, Appendix 4, and as described below, section 7.7.

7.7. Trial assessments

Trial assessments will be performed according to the Schedule of Procedures, Appendix 4. Where trial assessments identify any clinically significant incidental findings, these will be communicated to the participant's GP, with the participant's consent.

Trial assessments will be carried out according to trial specific guidelines.

Missed trial assessments or visits completed outside the visit window will not be reported as breaches where this is due to participant choice or a clinical decision, excursions will be documented, and trial statistician made of aware of discrepancies.

Medical history: focused medical history, taken from medical records and participant reporting, including the following information:

- History of chronic medical conditions related to inclusion and exclusion criteria
- Medication allergies

Review concomitant medications and therapies: taken from medical records and participant reporting at each visit.

Review of AEs: participants will be asked about the occurrence of any AEs since the previous visit, medical records will also be reviewed.

Severity of bronchiectasis: will be evaluated using the Bronchiectasis Severity Index and Medical Research Council dyspnoea score. Exacerbation assessment will use the EMBARC definition of exacerbation (Hill et al. 2017):

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 a clinician determines that a change in bronchiectasis treatment is required

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

Height & weight: as per trial guidelines.

Vital signs: Blood Pressure, pulse, temperature, oxygen saturation as per trial guidelines.

Pregnancy test (urine): will be carried out for women of childbearing potential (WOCBP), as described in section 6.2. See also notes in the exclusion criteria section 6.2, regarding contraception.

Electrocardiogram (ECG): as per trial guidelines, will be reviewed by a doctor prior to randomisation.

6-minute walk test: this test assesses the distance (in metres) an individual can walk in 6 minutes. The participant walks back and forth along a marked walkway of 20 meters at their own pace.

Spirometry: post bronchodilator spirometry at visits will be carried using American Thoracic Society/European Respiratory Society's guidelines. Forced Expiratory Volume (FEV1) forced vital capacity (FVC) and Forced Expiratory Flow at 25-75% (FEF25-75) will be measured.

Questionnaires: as per schedule of procedures.

Blood samples: collected, processed, and stored as per laboratory manual. Full blood count, urea and electrolytes, liver function tests will be analysed by the local NHS laboratory.

Samples will also be taken for trial outcomes. Additional blood samples will be collected for future biomarker and molecular microbiology research, dependent on participant consent, see section 7.2.1. A maximum of 100 ml of blood will be obtained at any visit.

Sputum sample: a sputum sample will be collected, processed, divided and stored as per laboratory manual. Participants will be asked to bring a spontaneous early morning sputum sample with them to visits from their second visit onwards. Where a participant is unable to produce a sputum sample at a visit a hierarchical approach to obtaining sputum samples will be used:

1. spontaneous sputum sample produced at the visit
2. spontaneous early morning sputum brought from home on the day
3. spontaneous early morning sputum brought in within the following 48 hours after the scheduled visit.
4. Induced sputum at sites able to perform induced sputum according to local protocols.

A sputum sample will be analysed locally for active neutrophilic inflammation at screening/baseline using NEATstik.

A sample will also be taken for trial outcomes. Additional sputum sample will be taken for future biomarker and molecular microbiology research, see section 7.2.1.

Nasal sample: brushing of the inferior turbinate will be performed to obtain nasal epithelial cells and the brush then placed into media for storage. Participants should not have a nasal brushing performed if they are receiving anticoagulants (Oral anticoagulants or warfarin) or

have a history of deviated nasal septum, base of skull fracture or severe epistaxis. If any of these are present the nasal brushing will be omitted.

Safety assessments: participants will be requested to attend an unscheduled visit if they experience a bronchiectasis exacerbation or any other safety concern.

7.7.1. Trial assessments, sub-study only

Pulse wave velocity: (PWV) is a non-invasive assessment to measure arterial stiffness, which is a surrogate marker that predicts cardiovascular events. PWV is defined as the speed of which the pulse wave travels along a length of artery, expressed in m/s, where higher values indicate higher cardiovascular risk (Oliver et al. 2003). PWV will be measured using the carotid-femoral technique, which is regarded as the gold-standard to measure large artery stiffness (Van Bortel et al. 2012). This is done using a device called the SphygmoCor® Xcel (AtCor Medical UK). The SphygmoCor® Xcel will calculate the distance divided by pulse transit time between carotid and femoral, provided by a tonometer over the carotid artery, and a blood pressure cuff over the femoral artery (Butlin et al. 2017).

Iontophoresis and full-field laser perfusion imaging (FLPI): iontophoresis is a non-invasive method that drives ionised medications across the skin with the use of a small amount of physiologically acceptable electric current (0.5 mA/cm² or less). This is performed using a set of electrodes, and one of which contains an ionised medication in a chamber. When electric current is applied, it will move the ionised medication from one electrode to another (Dhote et al. 2012). The medication can be a vasoactive compound such as acetylcholine (positive charged) or sodium nitroprusside (negative charged), which can then be used to assess vasodilation. An impairment in response to acetylcholine during iontophoresis implies endothelial dysfunction (Schonberger et al. 2006).

Full-field laser perfusion imaging (FLPI) measures the blood flow within the superficial microcirculation using laser speckle contrast imaging. FLPI uses a near infra-red laser diode (785nm) to illuminate the skin, and captures images of the superficial micro vessels at a depth of 300µm. The movement of red blood cells within the skin and tissues will cause the laser light to backscatter. The backscattered light will then get focussed through a limiting aperture and captured by a camera, creating speckle patterns. The speckle pattern is altered by movements of the red blood cells, and can be converted into a colourised image that represents blood flow within a region of tissue in real time (Adams 2014). FLPI can be combined with iontophoresis of vasoactive medications to provide an assessment of endothelial dysfunction.

Waist and hip measurement will be taken as the ratio of these measurements are useful predictors of heart attacks and strokes.

7.8. Long term follow-up assessments

Nil

7.9. Quality of life and symptom assessments

Quality of life will be evaluated using the QOL-B respiratory symptom scale (Quittner et al. 2015a) and the BIM.

Participants will complete a daily diary of symptoms BEST during the 28 day treatment period. This is completed at home either on paper and brought to the research visit or entered electronically via weblink or mobile application.

7.10. Withdrawal criteria

Participants are free to withdraw at any time and are not obliged to give reason(s). The CI, PI or delegate will make a reasonable effort to ascertain the reason(s), both for those who express their right to withdraw and for those lost to follow up, while fully respecting the individual's rights.

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 patient's well-being. A full explanation will be provided. If the trial is being conducted on an intention to treat basis, and the participant has been randomised and given one or more doses of IMP, s/he will be asked to complete trial visits as per the protocol, if the CI considers it appropriate. This would allow for an intention to treat analysis - but will be censored in the per-protocol analysis. Participants are free to refuse to do so. Withdrawn participants will not be prescribed IMP.

Those withdrawn, including those lost to follow-up, will be identified and a descriptive analysis of them provided, including the reasons for their loss, if known, and its relationship to treatment and outcome.

If a participant withdraws or is withdrawn, they have the right to ask for their data and samples held to be removed/disposed of, changed or deleted. This might not always be possible if it means the data/samples cannot be used to do the research, if this is the case, participants will be informed.

7.11. Storage and analysis of clinical samples

The collection, processing and storage of samples will be detailed in the Laboratory Manual. The analysis of samples will be detailed in the Laboratory Analytical Plan.

Blood and sputum samples will be obtained as per Laboratory Manual. Processing of blood for serum generation and cell isolation will be completed at sites, details of how processing will be carried out will be included in the Laboratory Manual. The resulting serum, sputum supernatant, whole sputum and cells will be stored at site.

All samples will be stored at sites and transferred to the UoD, Division of Molecular and Clinical Medicine, for analysis and storage either at the end of site's participation or sooner if site requests. Access to samples will be limited to members of the CI's laboratory team. Specimens will be registered with NHS Tayside Biorepository. Future use of those specimens will be made available and will be governed by the NHS Tayside Biorepository and access committee. Trial data may be released with samples.

It is the responsibility of the trial site to ensure that samples are appropriately labelled in accordance with the trial procedures to comply with the Data Protection Act 2018. Biological samples collected from participants as part of this trial will be transported, stored, accessed and processed in accordance with national legislation relating to the use and storage of human tissue for research purposes and such activities shall at least meet the requirements as set out in the 2004 Human Tissue Act and the 2006 Human Tissue (Scotland) Act.

7.12. End of trial

The end of trial at all sites is defined as last participant last visit (LPLV). The Sponsor, CI and/or the TSC 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, Research Ethics Committee (REC), Medicines and Healthcare products Regulatory Agency (MHRA) and National Health Service Research & Development (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 within 1 year of the end of the trial and will also be provided to the Sponsor and REC.

8. TRIAL TREATMENTS

8.1. Name, description and regulatory status of investigational medicinal products (IMPs) for open arms

	Treatment	Dosage, form and strength	Regulatory status
Arm 2	Disulfiram	400 mg oral tablets two 200mg tablets once daily	Not approved for the treatment of bronchiectasis. Has current marketing authorisation (MA) in the UK for use as an adjuvant in the treatment of drinking problems.
Arm 3	Dipyridamole	200mg oral prolonged/modified release capsule one capsule twice daily	Not approved for the treatment of bronchiectasis. Has current MA in the UK for use in secondary prevention of ischaemic stroke and transient ischaemic attacks and as an adjunct to oral anti-coagulation for prophylaxis of thromboembolism associated with prosthetic heart valves.
Arm 4:	Doxycycline	100mg oral capsule one capsule once daily	Not approved for the treatment of bronchiectasis. Has current MA in the UK for use in the treatment of a variety of infections caused by susceptible strains of Gram-positive and Gram-negative bacteria and certain other micro-organisms.

8.2. Name, description and regulatory status of IMP(s) for closed arms.

Nil, initial trial protocol, no treatment arms have been closed.

8.3. Product Characteristics

The reference safety information (RSI) is described in Section 9.4.

8.4. Accountability Procedures

The PI or delegated trial staff will maintain an accurate record of the receipt and dispensing of the IMP in a drug accountability log. Monitoring of drug accountability will be performed as per Sponsor Monitoring Plan.

8.5. Preparation and labelling of IMP

IMP stock will be taken from local hospital supply. The IMP will be labelled by delegated pharmacy staff, as per the pharmacy manual.

8.6. Drug supply and storage, open treatment arms

Where a site has difficulty obtaining the supply of medication for any of the treatment arms, the site will continue to recruit and randomise participants between the available treatment arms and usual care at that site.

	Treatment	Supply	Storage
Arm 2	Disulfiram	Generic drug, procured through site NHS pharmacy	As per summary of product characteristics (SmPC) conditions
Arm 3	Dipyridamole	Generic drug, procured through site NHS pharmacy	As per SmPC conditions
Arm 4	Doxycycline	Generic drug, procured through site NHS pharmacy	As per SmPC conditions

8.7. Dosage schedules, open treatment arms

	Treatment	Dosage schedule	Duration
Arm 2	Disulfiram	Two 200mg tablets once daily	28 days
Arm 3	Dipyridamole	One 200mg capsule twice daily	28 days
Arm 4	Doxycycline	One 100mg capsule once daily	28 days

8.8. Dosage modifications, open treatment arms

	Treatment	Dosage modifications
Arm 2	Disulfiram	Nil
Arm 3	Dipyridamole	Nil
Arm 4	Doxycycline	Nil

8.9. Known drug reactions and interaction with other therapies, open treatment arms

Treatment	Known drug reactions	Known drug interactions
Arm 2: Disulfiram	Disulfiram-alcohol reaction	<p>Alcohol - Disulfiram has several CYP450 enzyme interactions, which results in the interference of the metabolism of various drugs. Disulfiram inhibits CYP2E1 CYP1A2, and is activated by CYP3A4.</p> <p>Amitriptyline – increased disulfiram alcohol reaction</p> <p>Chlorpromazine – increased disulfiram alcohol reaction</p> <p>Benzodiazepines – enhanced sedative effect</p> <p>Phenytoin – increased phenytoin concentration and needs dose adjustment</p> <p>Warfarin – increased anticoagulant effect and needs dose adjustment</p> <p>Rifampicin – increased concentration due to reduced metabolism and renal excretion</p> <p>Metronidazole, Isoniazid and Paraldehyde were anecdotally found to increased risk of psychotic reaction</p>
Arm 3: Dipyridamole	<p>Very common, $\geq 1/10$</p> <p>Headache</p> <p>Dizziness</p> <p>Diarrhoea</p> <p>Nausea</p> <p>Common, $\geq 1/100$ to $< 1/10$</p> <p>Rash</p> <p>Angina pectoris</p> <p>Vomiting</p> <p>Myalgia</p> <p>Unknown drug reactions are detailed in the RSI.</p>	<p>Adenosine - Dipyridamole inhibits the uptake and degradation of adenosine in the blood vessels and cells. Adenosine use should be adjusted or avoided.</p> <p>Antiplatelets and anticoagulation – increases bleeding risk.</p> <p>Antihypertensives – increases risk of hypotension.</p> <p>Alcohol – increase the risk of hypotension.</p> <p>Cholinesterase Inhibitors in Myasthenia Gravis – worsens symptoms</p>

Arm 4: Doxycycline	<p>Common, $\geq 1/100$ to $< 1/10$</p> <p>Hypersensitivity Headache Nausea/vomiting Photosensitivity reaction, rash including maculopapular and erythematous rashes</p> <p>Uncommon, $\geq 1/1000$ to $< 1/100$</p> <p>Vaginal infection Dyspepsia</p> <p>Rare and unknown drug reactions are detailed in the RSI.</p>	<p>Antacids – may impair absorption of doxycycline</p> <p>Barbiturates, Carbamazepine, phenytoin, and primidone – may reduce exposure to doxycycline</p> <p>Warfarin - increased anticoagulant effect and needs dose adjustment.</p> <p>Oral contraception – few cases of breakthrough bleeding or pregnancy</p> <p>Alcohol – reduced doxycycline half-life</p> <p>Retinoids – increased risk of benign intracranial pressure</p> <p>Rifampicin - may reduce exposure to doxycycline</p>
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8.10. Concomitant medication

Details of all concomitant medications will be recorded on the trial eCRF on a concomitant medications log. Where possible participant's regular treatments received for the management of bronchiectasis should remain stable throughout the trial period.

8.11. Trial restrictions

	Treatment	Trial restrictions
All arms		To avoid contamination between the arms, participants who develop an exacerbation during the study and require antibiotic treatment should receive an antibiotic other than doxycycline. If participants are not able to safely avoid doxycycline during the trial period they should not be enrolled in the study.
Arm 2	Disulfiram	Alcohol Participants will be required to consent to not consuming alcohol for duration of treatment and at least 14 days after stopping treatment. Written advice will be provided to participants about how to avoid products containing alcohol during the trial period.
Arm 3	Dipyridamole	Participants should not commence new oral anticoagulant drugs during the trial period. Participants needing to initiate anticoagulants should withdraw from the study.
Arm 4:	Doxycycline	No restrictions. Exacerbations of bronchiectasis should be treated with an alternative antibiotic.
All	Nasal brushing	Participants should not have a nasal brushing performed if they are receiving anticoagulants (oral anticoagulants or warfarin) or have a history of deviated nasal septum, base of skull fracture or severe epistaxis. If any of these are present the nasal brushing will be omitted. This will not affect their participation in the trial.

WOCBP must be willing to have pregnancy testing prior to trial entry and at each research visit, unless the participant has been randomised to usual care.

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

In addition, WOCBP must be willing to use a form of a medically approved birth control method throughout the trial (and for minimum of 4 weeks after last dose), unless randomised to usual care, which include:

- combined (estrogen and progestogen containing) hormonal contraception associated with inhibition of ovulation:
 - oral
 - intravaginal
 - transdermal
- progestogen-only hormonal contraception associated with inhibition of ovulation:
 - oral
 - injectable
 - implantable
- intrauterine device (IUD)
- intrauterine hormone-releasing system (IUS)
- bilateral tubal occlusion
- vasectomised partner
- sexual abstinence, when this is in line with the preferred and usual lifestyle of the participant, abstinence is acceptable only as true abstinence. Periodic abstinence (e.g., calendar, ovulation, symptom-thermal, post-ovulation methods) and withdrawal are not acceptable methods of contraception.

8.12. Assessment of compliance with treatment

Adherence to trial drug will be checked by tablet counting at the last visit. Participants will also be asked at each visit regarding their adherence to taking the trial drug daily.

8.13. Name and description of each Non-Investigational Medicinal Product

Nil

9. PHARMACOVIGILANCE

9.1. Definitions

Term	Definition
Adverse Event (AE)	Any untoward medical occurrence in a participant to whom a medicinal product has been administered, including occurrences which are not necessarily caused by or related to that product.
Adverse Reaction (AR)	An untoward and unintended response in a participant to an IMP which is related to any dose administered to that participant.

	<p>The phrase "response to an IMP" means that a causal relationship between a trial medication and an AE is at least a reasonable possibility, i.e. the relationship cannot be ruled out.</p> <p>All cases judged by either the reporting medically qualified professional or the Sponsor as having a reasonable suspected causal relationship to the trial medication qualify as ARs. It is important to note that this is entirely separate to the known side effects listed in the SmPC. It is specifically a temporal relationship between taking the drug, the half-life, and the time of the event or any valid alternative etiology that would explain the event.</p>
Serious Adverse Event (SAE)	<p>A SAE is any untoward medical occurrence that:</p> <ul style="list-style-type: none"> • results in death • is life-threatening • requires inpatient hospitalisation or prolongation of existing hospitalisation • results in persistent or significant disability/incapacity • consists of a congenital anomaly or birth defect <p>Other 'important medical events' may also be considered serious if they jeopardise the participant or require an intervention to prevent one of the above consequences.</p> <p>NOTE: The term "life-threatening" in the definition of "serious" refers to an event in which the participant was at risk of death at the time of the event; it does not refer to an event which hypothetically might have caused death if it were more severe.</p>
Serious Adverse Reaction (SAR)	<p>An AE that is both serious and, in the opinion of the reporting Investigator, believed with reasonable probability to be due to one of the trial treatments, based on the information provided.</p>
Suspected Unexpected Serious Adverse Reaction (SUSAR)	<p>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:</p> <ul style="list-style-type: none"> • in the case of a product with a MA, this could be in the SmPC for that product, so long as it is being used within its licence. If it is being used off label an assessment of the SmPCs suitability will need to be undertaken. • in the case of any other IMP, in the investigator's brochure (IB) relating to the trial in question

NB: to avoid confusion or misunderstanding of the difference between the terms "serious" and "severe", the following note of clarification is provided: "Severe" is often used to describe intensity of a specific event, which may be of relatively minor medical significance. "Seriousness" is the regulatory definition supplied above.

Causality assessment

Unrelated	Where the AE is not considered to be related to the study drug: more likely. Information on drug withdrawal may be lacking or unclear
Possibly	Although a relationship to the study drug cannot be completely ruled out, the nature of the event, the underlying disease,

	concomitant medication, or temporal relationship make other explanations
Probably	The temporal relationship and absence of a more likely explanation suggest the event could be related to the study drug. Information on drug withdrawal may be available and if so the observed response to study drug withdrawal is considered clinically reasonable
Definitely	The known effects of the study drug or its therapeutic class, or based on challenge testing, suggest that the study drug is the most likely cause. Information on drug withdrawal is usually available and the observed response to study drug withdrawal is considered clinically reasonable and has a plausible temporal relationship to study drug exposure

9.2. Operational definitions for (S)AEs

Worsening of bronchiectasis or bronchiectasis exacerbations during the trial will not be classed as an AE but is defined as an outcome. However, hospitalisations resulting from worsening of bronchiectasis will be recorded as AEs and reported as SAEs.

An abnormal laboratory finding, that requires medical intervention by the investigator, or a finding judged by the investigator as medically significant, will be reported as an AE. If clinical sequelae are associated with a laboratory abnormality, the diagnosis or medical condition will be reported (e.g. renal failure, haematuria) not the laboratory abnormality. Abnormal laboratory values that are not, in the investigator's opinion, medically significant and do not require intervention will not be reported as AEs.

A non-clinically significant, in the opinion of the investigator, worsening of a pre-existing condition during the trial will not be classed as an AE. Pre-specified elective hospitalisations for treatment planned prior to randomisation will not be considered as an AE. However, any AEs occurring during such hospitalisations will be recorded.

9.3. Recording and reporting of SAEs, SARs AND SUSARs

All AEs will be recorded on the AE Log in the eCRF. Details of AEs will be recorded in the medical record. AEs will be assessed for severity by the PI. AEs will be recorded from the time a participant consents to join the trial until the participant's last trial visit. Any SUSAR, that the investigator becomes aware of, will be reported to the Sponsor irrespective of how long after IMP administration the reaction has occurred.

Assessment of severity of AEs:

Mild: An event that is easily tolerated by the participant, causing minimal discomfort, and not interfering with everyday activities.

Moderate: An event that is sufficiently discomforting to interfere with normal everyday activities.

Severe: An event that prevents normal everyday activities.

An AE may be classified as a SAE or AR.

Participants with unresolved AEs/SAEs at end of trial will be followed up until 30 days after participant's last visit. SUSARS will be followed until resolution, where a participant agrees to this.

The CI, PI or delegate will ask about the occurrence of AEs and hospitalisations at every visit during the trial. SAEs will be submitted on an SAE form to the Sponsor Pharmacovigilance Section via the online Tayside Pharmacovigilance System within 24 hours of becoming aware of the SAE. Site PIs will also notify the CI when submitting an SAE.

The evaluation of expectedness will be made based on the knowledge of the reaction and the RSI see Section 9.4. The Sponsor will make the definitive assessment on expectedness for the purposes of SUSAR reporting.

The Sponsor is responsible for reporting SUSARs to the Competent Authority, and the REC. Fatal or life-threatening SUSARs will be reported within 7 days and non-fatal and non-life threatening SUSARs within 15 days.

Reporting of safety data to the funders will be as detailed in the funding agreement.

9.4. Reference Safety Information, open treatment arms

The current version, of the RSI will be held in the TMF - specifically in Pharmacy Site File, Investigator Site File (ISF) and Sponsor File. The RSI will be reviewed at least annually and where there have been any changes to the Reference Safety Information which may impact on the trial the protocol will be reviewed and a substantial amendment submitted for regulatory approvals.

	Treatment	Reference Safety Information
Arm 2	Disulfiram	Summary of Product Characteristics, Section 4.8
Arm 3	Dipyridamole	Summary of Product Characteristics, Section 4.8
Arm 4:	Doxycycline	Summary of Product Characteristics, Section 4.8

Mitigation of risks is detailed in Appendix 1.

9.5. Responsibilities

CI/PI or delegated staff:

- Checking for AEs and ARs at all visits.

CI/PI or medically qualified delegate:

- Confirmation of eligibility criteria.
- Using medical judgement in assigning seriousness, causality and whether the event/reaction was related.
- Ensuring that all SAEs are recorded and reported to the sponsor within 24 hours of becoming aware of the event and provide further follow-up information as soon as available.

CI:

- Central data collection of AEs, ARs, SAEs, SARs and SUSARs according to the trial protocol onto a database.
- Ensuring that AEs and ARs are recorded and reported to the sponsor in line with the requirements of the protocol.

- Clinical oversight of trial participant safety, including an ongoing review of the risk/benefit.
- Immediate review of all SUSARs.
- Assigning Medical Dictionary for Regulatory Activities (MedDRA) or Body System coding to all SAEs and SARs.
- Periodically reviewing overall safety data to determine patterns and trends of events, or to identify safety issues, which would not be apparent on an individual case basis.
- Preparing the clinical sections and final sign-off of the Development Safety Update Report.
- Reporting safety information to funder as per contract.

Sponsor (UoD/NHS Tayside):

- Expedited reporting of SUSARs to the MHRA and REC within required timelines.
- Checking for (annually) and notifying PIs of updates to the Reference Safety Information for the trial.
- Preparing standard tables and other relevant information for the DSUR in collaboration with the CI and ensuring timely submission to the MHRA and REC.

Trial Steering Committee (TSC):

In accordance with the TSC Charter, the TSC will periodically consider recommendations from the DMC following their review of safety data and make decisions on early termination or continuation of recruitment to individual treatment arms and the overall trial.

Data Monitoring Committee (DMC):

In accordance with the DMC Charter, the DMC will periodically review overall safety data to determine patterns and trends of events, and identify potential safety issues, making recommendations to the TSC.

9.6. Notification of deaths

All deaths occurring during the trial, will be reported to the sponsor irrespective of whether the death is related to disease progression, the trial drug or an unrelated event. Deaths will be reported to Sponsor as SAEs as per Section 9.3.

9.7. Pregnancy reporting

Pregnancy itself 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 within 24 hours of becoming aware of the pregnancy and the outcome 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.

9.8. Overdose

An overdose with associated AEs is recorded as the AE diagnosis/symptoms on the relevant AE section in the eCRF. Any dose administered other than prescribed dose for that participant will be reported to the Sponsor as a protocol breach.

If an overdose of trial drug occurs during the trial, then the Investigator or other site personnel will inform the appropriate sponsor representatives immediately, or no later than 24 hours after becoming aware of it. The designated sponsor representative will work with the Investigator to ensure that all relevant information is provided to the sponsor's Pharmacovigilance Committee.

	Treatment	Definition	Treatment
Arm 2	Disulfiram	Exceeding the trial dose of 400mg per day.	Treatment should be symptomatic, and observation is recommended. Supportive therapy should be available, and measures may be necessary to counteract hypotension. Gastric lavage and/or activated charcoal may be considered in cases of disulfiram overdose. Severe vomiting might occur requiring administration of intravenous fluids.
Arm 3	Dipyridamole	Exceeding the trial dose of 400mg per day	Symptomatic therapy is recommended. Administration of xanthine derivatives (e.g. aminophylline) may reverse the haemodynamic effects of dipyridamole overdose. ECG monitoring is advised in such a situation.
Arm 4:	Doxycycline	Exceeding the trial dose of 100mg per day	Discontinue medication. Gastric lavage plus appropriate supportive treatment may be indicated.

9.9. Reporting urgent safety measures

The PI or other trial physician will take appropriate immediate urgent safety measures 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.

9.10. The type and duration of the follow-up of participants after adverse reactions.

All ARs will be recorded as per section 9.3. Where ARs occur, assessment of clinical condition and appropriate treatment will be instigated by a delegated doctor and will continue until the symptoms resolve or the condition stabilises.

9.11. Development safety update reports

The 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.

The DSUR and reports of SUSARs will be sent to REC by the Sponsor Pharmacovigilance Section. Any other safety reports, for example, reports of a DMC, will be sent by the CI to REC, with a Safety Report Form, and to the Sponsor.

10. STATISTICS AND DATA ANALYSIS

10.1. Sample size calculation

Given the nature of the trial, there is no overall sample size target, but the numbers of participants required for each treatment arm are:

	Treatment	n	Open/closed to recruitment
Arm 1	Usual care	Randomisation will continue to end of trial	Open
Arm 2	Disulfiram	42	Open
Arm 3	Dipyridamole	42	Open
Arm 4	Doxycycline	42	Open

Sample size calculations have been performed using the “Superpower” package in R version 4.2.1 and verified using simulations.

The primary endpoint is sputum NE at day 28. The standard deviation of log₁₀-transformed NE is estimated to be 0.5, using unpublished data from the ORBIT4 trial. A “moderate” standardised difference (Cohen, 1988) of 0.5 between the control and each treatment arm is targeted, which equates to a log₁₀-difference of 0.25, i.e. a reduction in NE of approximately 44% vs control after 28 days of active treatment. The DPP1 inhibitor Brensocatib achieved >90% reduction in NE, so the targeted difference is realistic.

Aiming to recruit the number of participants per treatment arm as above (to allow for up to 10% dropout), with concurrent equal randomisation to the shared control arm, the trial would have >80% power to detect such a difference at the 1-sided 10% significance level. This assumes adjustment for up to 4 covariates in the ANCOVA model, with a modest R² value of approx. 0.1. In reality, the R² value should be larger than this (based on ORBIT-4), so power should be closer to 90% (see Table 10.1.1) (Haworth et al. 2019).

Table 10.1.1. Power, given R² value of covariates, to detect a standardised difference in log₁₀-transformed NE at day 28 of 0.5 between each treatment arm and the shared control at the 1-sided 10% significance level.

R ² value of covariates (with up to 4 covariates)	Power
0.1	81.8%
0.2	85.1%
0.3	88.5%
0.4	91.9%

The design uses the rationale of randomised phase II screening designs in oncology (Rubinstein et al. 2005). By setting the type I error rate to 10% (higher than the conventional

5% for phase III trials but widely accepted in phase II) and targeting a moderate treatment effect size, the required sample size is restricted while maintaining good power. This allows us to quickly and efficiently evaluate multiple treatment options, screening out those that appear suboptimal, and taking forward any showing a sufficiently promising efficacy signal for confirmation in a larger randomised trial.

The analysis of each treatment arm will be performed separately. Treatment arm participants will only be compared to contemporaneously recruited control arm participants. Type I error rate is controlled at 10% for each pairwise comparison.

To aid recruitment, after completing the 56 day follow-up period, patients may be rescreened and re-randomized if they are willing to participate in the trial again. Re-randomisation will be performed independently, in the same way as for initial randomisations. (Kahan et al. 2015). There may be an increase in power using this approach, but with few patients expected to opt for re-randomisation, and multiple active arms decreasing the likelihood of re-randomisation to the same pairwise comparison, any benefit is expected to be small.

10.1.1. Sample size calculation, sub-study

The sub-study is being carried out for exploratory outcomes only and no sample size calculation is required.

10.2. Planned recruitment rate

Recruitment to the first 3 treatment arms and contemporaneously randomised control arm participants is expected to take approximately 22-30 months across 10 recruiting sites. As treatment arms are added recruitment will continue.

10.3. Statistical analysis plan

The primary analysis will be performed on an intention-to-treat basis. Analysis of covariance (ANCOVA) will be used to compare the log₁₀-transformed NE between the control and each treatment arm after 28 days, with baseline log₁₀-transformed NE included as a covariate in addition to terms for trial arm, minimisation factors and relevant baseline prognostic variables. Missing data will be dealt with using multiple imputation, assuming missing-at-random is an appropriate assumption; methods exploring other mechanisms may be performed as appropriate. The estimated difference between the arms in log₁₀-transformed NE will be presented along with the corresponding 80% confidence interval and 1-sided p-value. Analysis makes a working independence assumption. To account for the re-randomisation of patients, cluster-robust standard errors will be computed (Kahan et al. 2021)

NE over the entire study period will be compared between arms using linear mixed effects models, including fixed effect terms for trial arm, stratification factors and relevant baseline prognostic variables, and a random intercept participant effect. Quality of life outcomes will be compared between arms using a similar approach.

Analysis of time to first exacerbation will be conducted using a Cox regression model incorporating trial arm, stratification factors and relevant baseline prognostic variables. The hazard ratio, 80% CI and 1-sided p-value associated with study arm will be obtained from this model, using cluster-robust standard errors. A Kaplan-Meier plot will be used to illustrate the relative time to first exacerbation in each arm and contemporaneous controls, with

proportion of patients exacerbation-free after 28 and 56 days and associated 80% CIs provided.

Safety will be assessed by tabulating AEs that occurred in at least 10% of the safety population participants in either arm; these will be further broken down by severity. Treatment compliance (including number of expected doses taken/missed, number of dose reductions (if applicable), reasons for missed doses/dose reductions, etc) will be summarised for each treatment arm.

Full details, including planned sensitivity analyses and the approach for handling missing data, will be provided in a separate Statistical Analysis Plan.

10.3.1. Statistical analysis plan, sub-study

The results from the sub-study assessments will be combined with results from separate trials, conducted in people with bronchiectasis, not connected with this protocol for analysis. A separate statistical analysis plan will be prepared for the sub-study analysis.

10.4. Interim analysis and criteria for the premature termination of treatment arms or the trial

There will be no interim analyses considering early stopping for efficacy or futility in any treatment arms.

There will be an early review of safety and treatment compliance data by the DMC in each treatment arm. This will take place after the 10th randomised participant to the treatment arm has reached 28 days post-randomisation. Based on this review, the DMC will make recommendations to the TSC as to whether recruitment to the treatment arm should stop, continue as planned, or whether any modifications should be considered (e.g. dosing).

10.5. Economic evaluation

No economic evaluation will be performed.

11. DATA MANAGEMENT

11.1. Data collection tools and source document identification

The PI or delegate will maintain source documents for each participant in the trial, consisting of hospital medical records containing demographic and medical information, laboratory data, electrocardiograms, trial questionnaires and the results of any other tests or assessments. The questionnaires will be completed by the participants and act as source data, the completed form will be filed in the ISF. All trial data relevant to a participant's general medical history will be recorded in the medical record. The medical record will be flagged to state that the participant is participating in the AIR-NET trial.

An eCRF, using Castor Electronic Data Capture (EDC) system, will be provided by TCTU. The trial system will be based on the protocol for the trial. Development and validation of the trial database and quality control will be done according to TCTU procedures. 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.

Participants will also have the option to input daily BEST diary data directly into either the Castor electronic Patient Reported Outcomes - Web version or the Castor Connect App which can be downloaded and installed in the participant's mobile or tablet devices. Electronic Patient Reported Outcomes data collected via Web email and Castor Connect App will automatically be stored and synced to the Castor EDC.

The PI may delegate eCRF data entry but is responsible for completeness, plausibility, and consistency of the eCRF. Delegated trial staff will enter the data required by the protocol into the eCRFs following training in the definitions and methods used in completing the eCRF. Any queries will be resolved by the PI or delegated member of the trial team. On completion of data collection, the PI must certify that the data entered are complete and accurate.

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

All electronic data will be stored on secure UoD or cloud-based servers which have restricted access and have disaster recovery systems in place.

11.2. Data handling and record keeping

The database is managed in line with all applicable principles of medical confidentiality and UK law on data protection, namely, the Data Protection Act 2018. The Data Controller will be the UoD and the Data Custodian will be the CI.

Development and validation of the trial database, quality control and data extraction will be managed by TCTU. Details will be documented in the Data Management Plan.

11.3. Access to Data

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 or inspection, the CI and/or PI will allow the Sponsor, representatives of the Sponsor or regulatory authorities direct access to all trial records and source documentation.

Anonymised trial data will be kept under the control of the CI for future research use within the UoD and with other research collaborators (commercial and non-commercial). Access to the data will be as described in section 13.7.

11.4. Archiving

Archiving of trial documents will be in compliance with Sponsor Standard Operating Procedures. Medical records will be maintained in compliance with local NHS policy on retention of medical records. The CI will be responsible for arranging the archiving of the TMF and ensuring that research data is archived in a way that will permit accurate reconstruction of the research. Sites will be responsible for archiving local trial records including the ISF and Pharmacy Site File. Sponsor will be responsible for archiving the Sponsor file.

12. MONITORING, AUDIT & INSPECTION

12.1. Monitoring

A trial risk assessment will be carried out by the Sponsor prior to Sponsorship approval being granted. The Sponsor will determine the appropriate extent and nature of monitoring for the trial and will delegate monitoring to appropriately qualified and trained monitors. A Monitoring Plan will be developed by the Sponsor based on the trial risk assessment which will include on site and/or remote monitoring. The Monitoring Plan will be reviewed regularly using a risk-based approach and updated as required. The Monitoring Plan will detail the procedures and anticipated frequency of monitoring and processes reviewed. Sites must have access to source data for purposes of remote monitoring and assist the Sponsor in monitoring of the trial. In recognition that source data may come from different sources at each site, sites shall ensure that a source data identification list is supplied to the Monitoring Team in advance of any monitoring review and ensure this data is available on the agreed date and time to facilitate the review.

13. ETHICAL AND REGULATORY CONSIDERATIONS

13.1. Research Ethics Committee (REC) review & reports

Before the start of the trial, approval will be sought from an independent REC for the trial Protocol, Informed Consent Form, and other relevant documents.

Substantial amendments that require review by REC will not be implemented until the REC grants a favourable opinion for the trial.

All correspondence with the REC will be retained in the TMF.

A progress reports will be submitted to the REC according to REC approval conditions. It is the CI's responsibility to produce the REC reports as required.

The CI will notify the REC of the end of the trial. If the trial is ended prematurely, the CI will notify the REC, including the reasons for the premature termination. The CI will submit a final report with the results, including any publications/abstracts, according to REC approval conditions.

A copy of all REC reports will be submitted to the Sponsor.

13.2. Peer review

This trial has been funded by LifeArc who have reviewed the grant application. The trial has also been peer reviewed by EMBARC. The protocol has been reviewed and approved by the Sponsor Committee.

Resulting publications will be reviewed by the referees of the journal to which the paper will be submitted.

13.3. Public and Patient Involvement

Input from patients will be incorporated into each phase of the project through engagement with the European Lung Foundation patient advisory group, part of EMBARC.

Patient representatives will be incorporated into the trial steering committee, the TMG and will advise on all aspects of study design, implementation and dissemination. The patient

advisory group has been operating since 2015 and is a group of international bronchiectasis patients supported by the European Lung Foundation and inputs into clinical research across the EMBARC programme.

The patient representatives have given input into the participant facing documents and aspects of trial design relating to trial participation and burden.

13.4. Regulatory Compliance

The trial will not commence until a Clinical Trial Authorisation is obtained from the MHRA and favourable REC opinion. The protocol and trial conduct will comply with the Medicines for Human Use (Clinical Trials) Regulations 2004 and any relevant amendments.

Before any site can enrol participants into the trial, the CI, PI, or delegate will ensure that appropriate approvals from participating organisations are in place.

For any amendment to the trial, the CI, PI, or delegate, in agreement with the sponsor, will submit information to the appropriate body for them to review and issue approval for the amendment. The CI, PI or delegate will work with sites so they can put the necessary approvals and arrangements in place to implement the amendment to confirm their support for the trial as amended.

13.5. Protocol compliance

Prospective, planned deviations or waivers to the protocol are not allowed, e.g., it is not acceptable to enrol a participant if they do not meet the eligibility criteria or restrictions specified in the trial protocol. Trial staff will not implement deviations to the protocol except where necessary to eliminate an immediate hazard to trial participants.

Accidental protocol breaches can happen at any time. They will be adequately documented on the relevant forms and reported to the CI and Sponsor using the TASC Breach Reporting Form. If there is a breach of the protocol, the nature of and reasons for the breach will be documented in the trial Breach Log. Breaches from the protocol which are found to frequently recur are not acceptable, will require immediate action and could potentially be classified as a serious breach.

13.6. Notification of Serious Breaches to GCP and/or the protocol

A “serious breach” is a breach which is likely to affect to a significant degree –

- a) the safety or physical or mental integrity of the participants of the trial; or
- b) the scientific value of the trial

The sponsor will be notified immediately of any case where the above definition applies during the trial conduct phase. The sponsor of a clinical trial will notify the licensing authority in writing of any serious breach of

- a) the conditions and principles of GCP in connection with that trial; or
- b) the protocol relating to that trial, as amended from time to time.

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

If a breach necessitates a subsequent protocol amendment, this will be submitted as per section 13.10.

13.7. Data protection and participant confidentiality

The CI and trial staff will comply with the requirements of GDPR and the UK Data Protection Act 2018 or any subsequent amendment or replacement thereof with regard to the collection, storage, processing and disclosure of personal data and will uphold the principles of GDPR in Article 5.

The CI and trial staff will also adhere to the NHS Scotland Code of Practice on Protecting Participant Confidentiality or local 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 usernames and passwords. Age, gender, and ethnicity will be the only personal identifiable details held on Castor EDC system. Date of Birth will be held on the TRuST system to allow for the identification of participants where emergency unblinding is required.

Personal data or data concerning health will not be released without the existence of a legal basis for processing under Articles 6 and 9 of GDPR, such as official authority 6(1)e or substantial public interest 9(2)g. 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. In the event that data are shared with collaborators or groups wishing to undertake further analysis, collaborators will not have access to personal identifiable details other than those held on the EDC system. Pseudonymised participant data will also be available to interested parties after publication of the final report upon reasonable written request to the CI and subsequent approval.

The transfer of data to collaborators or for use in further research will be as described in the Clinical Research Agreement.

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

13.8. Financial and other competing interests for the Chief Investigator, PIs at each site and committee members for the overall trial management

The CI has worked as an advisory to the funder, LifeArc.

Additional disclosure information, if any, will be collected at site initiation and documented in the site file. Members of the TSC and DMC will complete a competing interest form, which will be held in the TMF

13.9. Indemnity

The UoD and Tayside Health Board are Co-Sponsoring the trial.

Insurance. – The UoD will obtain and hold Clinical Trials indemnity 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 UoD 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.

Other participating sites will maintain membership of a scheme similar to CNORIS.

13.10. Amendments

The closing of treatment arms and the addition of new treatment arms will require an amendment to the protocol.

Amendments to the protocol will be conducted in compliance with Sponsor Standard Operating Procedures. The decision to amend the protocol will lie with the CI after consultation with the TMG, and trial statistician. The TSC will also be consulted on any major amendments. The CI will seek Sponsor approval for any amendments to the Protocol or other approved trial documents. The Sponsor will decide whether an amendment is substantial or non-substantial. The CI will be responsible for submitting the amendment to the appropriate regulatory authorities and communicating amendments to sites. 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 appropriate site approvals. The amendment history will be detailed in an Amendment Log.

13.11. Post trial care

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 drug will be made by the trial team or Sponsor.

13.12. Access to the final trial dataset

The CI and Trial Statistician will have access to the final trial dataset. Access to the final trial dataset to others will be approved by the CI. See also section 13.7.

14. DISSEMINATION POLICY

Details of the trial and clinical trial final report will be published on ISCTRN Registry, no later than 12 months after the end of trial. Trial results will be available to the public via the ISCTRN registry. 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 participant representatives will be invited to suggest further dissemination activities for patients.

Participants in the trial will be notified of the results via a Results Letter.

14.1. Authorship eligibility guidelines and any intended use of professional writers

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

Authorship and the publication will be defined and developed by the TSC and the site investigators. An inclusive approach will be taken with either named or group authorship (e.g., “on behalf the AIR-NET Investigators”). In the case of group authorship all contributing participants will be named in (for example) a supplementary appendix. Named authors will be expected to meet authorship criteria set out by the International Committee of Medical Journal Editors.

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16. APPENDICIES

16.1. Appendix 1-Risk

<p>Risks associated with trial interventions</p> <p><input type="checkbox"/> A ≡ Comparable to the risk of standard medical care</p> <p><input checked="" type="checkbox"/> B ≡ Somewhat higher than the risk of standard medical care</p> <p><input type="checkbox"/> C ≡ Markedly higher than the risk of standard medical care</p>
<p>Justification:</p> <p>These repurposed therapeutics have established real-world evidence based on their safety profile, side effects, pharmacokinetics and pharmacodynamics, drug–drug interactions, and experience in different patient populations.</p>
<p>Outline any other processes that have been put in place to mitigate risks to participant safety (e.g. DMC, independent data review, etc.)</p> <p>There will be an early review of safety and treatment compliance data by the DMC in each treatment arm. This will take place after the 10th randomised participant to the treatment arm has reached 28 days post-randomisation. Based on this review, the DMC will make recommendations to the TSC as to whether recruitment to the treatment arm should stop, continue as planned, or whether any modifications should be considered (e.g. dosing).</p>
<p>Outline any processes (e.g. IMP labelling +/- accountability +/- trial specific temperature monitoring) that have been simplified based on the risk adapted approach.</p> <p>Nil</p>

Potential or identified risk of clinical significance	Summary of data/rationale for risk	Mitigation Strategy
Doxycycline		
Photosensitivity	Photosensitivity erythematous rash was observed in individuals undergoing doxycycline for infection treatment or malarial prophylaxis and tends to resolve upon discontinuation. This is thought to be due to ultraviolet (UV)-A radiation, mediated by mitochondrial toxicity and oxidative stress. Individuals with lighter complexion or on higher doses may be more prone to photosensitivity. Incidence for photosensitivity is highly	-Participants randomised to the doxycycline arm are advised to avoid sun exposure, wear protective clothing and use broad spectrum sunscreen (against UV-A and UV-B radiation)

	variable depending on location and population, ranging between 7.3-21.2% of individuals. (Tan et al. 2011).	
Gastrointestinal Disorder	People on doxycycline commonly experience gastrointestinal symptoms which are generally mild to moderate, including nausea, abdominal pain, vomiting, and diarrhoea. Doxycycline may also cause oesophageal inflammation or ulcers. It is considered that long oesophageal transit time and dehydration contributes to nausea and mucosal inflammation related to doxycycline. (Tan et al. 2011).	- Participants are advised to ingest the pill in an upright position, with plenty of fluid, and avoid taking before bedtime.
Disulfiram 400mg		
Disulfiram-Ethanol Reaction	Disulfiram-Ethanol Reaction (DER) mostly occurs in the first 12 hours after administration. Although severity of the DER is related to the disulfiram dose and ethanol intake, it was also seen in cases with blood alcohol as low as 10mg/100ml. A study found 19% of patients on disulfiram experienced DER while using alcohol-based hand sanitizer. (Lanz et al. 2023).	- Participants randomised to Disulfiram will be advised to abstain from alcoholic beverages or the use of any alcohol-based products during intervention - Proper ventilation during administration may alleviate DER when using alcohol based hand sanitizer
Hepatotoxicity	Carbon disulfide is released during hepatic disulfiram metabolism, and can cause hepatocellular degeneration or necrosis. Hepatotoxicity have previously manifested in the form of asymptomatic transaminitis to rare fulminant hepatic failure. This may be attributed to pre-existing alcoholic liver condition, but also found in 25% patients with	- Regular clinical and laboratory monitoring for hepatotoxicity

	no previous alcohol misuse. Most of these effects are reversible when Disulfiram is stopped. (Lanz et al. 2023).	
Dipyridamole		
Headache	<p>Headaches are a well-known transient side effects of dipyridamole, due to its vasodilatory properties. This often leads to discontinuation, as seen in 8% of patients who were enrolled into the dipyridamole arm in the Second European Stroke Prevention (ESPS2) Trial.</p> <p>A bioequivalence trial comparing two different formulations of dipyridamole involving mostly young healthy adults found that 72% of volunteers reported at least one episode of headache over the course of two weeks. This bioequivalence trial also found that the volunteers are more likely to experience headache in the first week, 2-3 hours after the morning dose, and all volunteers recovered fully from the headaches regardless of severity. (Theis, Deichsel, and Marshall 1999)</p>	- Participants should be informed that they may experience transient headaches while initiating dipyridamole, and reassurance that it will improve with time.
Bleeding	<p>As an antiplatelet, patients may be at a higher risk of bleeding complications. Adding dipyridamole to anticoagulation increases the risk of bleeding by about 50% (Massel and Little 2013).</p> <p>However, for patients not on anticoagulation, this risk was not seen in the large clinical trials performed in patients who had a Stroke or Transient Ischaemic Attack (TIA). In</p>	<p>- Participants who are on dual antiplatelet, any anticoagulant, or any coagulopathy should not be randomised to dipyridamole. Participants who are already on Aspirin are safe to be recruited.</p> <p>- Participants who recently had any gastrointestinal bleeding, head trauma, or major operation within 30 days should not be randomised to dipyridamole</p>

	ESPS2, patients who were randomised to placebo (n=1649) and dipyridamole alone (n=1654) reported similar events for bleeding any site, 74 and 77 respectively. ESPS2 also found that the bleeding risk between Aspirin (n=1649) and Aspirin plus Dipyridamole (n=1650) are similar at 135 and 144 events respectively (Diener et al. 1996).	
Hypotension	Dipyridamole is a potent vasodilator and the intravenous form was used in cardiac investigations. Oral dipyridamole is unlikely to induce hypotension, and was well tolerated even in patients with Escherichia coli endotoxin induced endotoxemia. It remains a theoretical risk and should be used with caution (Ramakers et al. 2011).	- Participants with severe coronary artery disease, recent myocardial infarct, or left ventricular systolic dysfunction can be recruited if their condition is stable.
Neutrophilic Inflammation Attenuation		
Infection	Theoretically, neutrophilic inflammation attenuation may increase the number of infections. This is however not commonly observed in clinical trials that used either disulfiram, dipyridamole or doxycycline in 1 month. The perceived risk is small.	- Regular clinical and laboratory monitoring for infection - An independent DMC is in place to periodically evaluate clinical trial safety data.

16.2. Appendix 2 - Trial management / responsibilities

Responsibilities will be detailed in the co-sponsorship and participating site agreements.

16.2.1. Participant registration/randomisation procedure

TCTU TRuST web-based randomisation system will be used.

Sites will be provided with a randomisation guide detailing the web-based randomisation system process. Prior to recruitment, individuals delegated this task will be given an individual username and password upon completion of training.

16.2.2. Data management

Data management will be overseen by TCTU Data Management Team.

Local sites will be expected to enter data directly on to the eCRF. Worksheets will be provided to facilitate this process, but their use is not mandatory. Worksheets, where used, will not be used for monitoring purposes.

All data from participants should be entered on the eCRF within 7 days of the last data collection point for that participant.

Data queries will be generated by the Data Management Team and should be addressed within 2 weeks.

16.2.3. Preparation and submission of amendments

TCTU Trial Management Team will be responsible for working with the CI to submit any amendments.

16.2.4. Preparation and submission of Annual Safety Report/Annual

TCTU Trial Management Team will be responsible for liaising with the CI to submit REC annual reports. The Sponsor Pharmacovigilance Team will be responsible for liaising with the CI to submit DSURs.

16.2.5. Data protection/confidentiality

The CI and trial staff will comply with the requirements of the Data Protection Act 2018, GDPR and the Data Protection Act 2018, or any subsequent amendment or replacement thereof regarding the collection, storage, processing and disclosure of personal data. The CI and trial staff will also adhere to the NHS Scotland Code of Practice on Protecting Participant Confidentiality or local equivalent.

16.2.6. Trial documentation and archiving

Archiving trial site data will be the responsibility of individual sites. Payment for archiving will be provided as per site agreement.

16.3. Appendix 3 – Authorisation of participating sites

16.3.1. Required documentation

The following data should be made available to TCTU Trial Management Team prior to site initiation:

- PI CV, signed and dated within the last 2 years
- PI GCP certificate
- Protocol signature page, signed and dated by PI
- Copy of signed Participating Site Agreement
- Copy of NHS R&D confirmation of capacity and capability
- Copy of the Delegation Log.

The following data should be made available and held within the ISF/Pharmacy Site File prior to site initiation:

- CV, signed and dated for all trial staff listed on Delegation Log
- GCP certificate for all trial staff listed on Delegation Log

16.3.2. Procedure for initiating/opening a new site

Site Initiation will be performed by Monitors and TCTU Trial Management Team and may be on site and/or remote.

TCTU Trial Management Team will initiate release of trial drug to the site after NHS R&D confirmation of capacity and capability.

16.3.3. Principal Investigator responsibilities

The PI's legal responsibilities will be listed in the Participating Site Agreement. A summary is given below:

- Attendance at the site initiation meeting
- Training of new members of trial staff in the protocol and its procedures,
- Ensuring that the ISF is accurately maintained
- Dissemination of important safety or trial related information to all stakeholders within their site
- Safety reporting within the required timelines
- Ensuring data entry to eCRF and responses to data clarification queries are completed within the required timelines
- Certify data entered on eCRF is correct and complete
- Ensuring any trial staff coming into contact with participants have the appropriate Personal Protective Equipment and training in its use
- Archiving of site trial data

16.4. Appendix 4 – Schedule of Procedures

	Visit 1 Screening	Visit 2 Baseline	Visit 3	Visit 4	Visit 5	Visit 6	Safety visit
	Days -35 to 0	Day 0	Day 7	Day 14	Day 28	Day 56	If required
Informed consent	X						
Eligibility check	X	X					
Demographics	X						
Medical history	X						
Concomitant medications	X	X	X	X	X	X	X
Physical examination	X						
Height & weight	X						
Record AEs		X	X	X	X	X	X
Record exacerbations		X	X	X	X	X	X
BP, pulse, temp, O ₂ stats	X	X	X	X	X	X	X
ECG	X						
6 minute walk test		X			X		
Post bronchodilation spirometry	X	X	X	X	X	X	X
QoL-B	X	X	X	X	X	X	
BIM baseline		X					
BIM follow-up			X	X	X	X	
BEST diary		X	X	X	X		
Full blood count, urea and electrolytes, liver function tests	X			X	X	X	X
Research blood sample for endpoint analyses	X	X	X	X	X	X	X
Sputum sample for screening eligibility (NEATstik) ^a	X						
Research sputum sample for endpoint analyses	X	X	X	X	X	X	X
Nasal sample		X			X		
Pregnancy test, if required	X	X	X	X	X		
Randomisation		X					
Dispensing of trial drugs		X					
Drug accountability					X		
Sub-study only							
Pulse wave velocity (PWV)		X			X	X	
Full-field Laser perfusion imaging (FLPI)		X			X	X	
Waist & Hip measurement		X					

^a If NEATSTIK is not available for screening, a frozen sputum sample will be shipped to the central laboratory in Dundee where the immunoassay will be performed and used to confirm eligibility.

Missed trial assessments or trial medication, or visits completed outside the visit window, will not be reported as breaches, where this is due to participant choice or a clinical decision.

Visits should be every as per schedule wherever possible. If a participant is unable to attend on the day of visit a delay of up to 2 days is accepted.

16.5. Appendix 5 – Safety Reporting Flow Chart

Activity	Responsibility	Timing	Comments
Review medical records and questioning of participant for evidence of AEs at all visits.	Trial staff	All visits	Recorded on eCRF system.
Review of recorded AEs for causality and seriousness	PI (or delegate)	Within 24 hours of recording	Recorded on eCRF and/or medical records.
Reporting SAEs - All SAEs need to be assessed and signed off by the PI or delegated doctor.	PI (or delegate)	Within 24 hours of becoming aware of SAE	Reported via the online Tayside Pharmacovigilance system
Reviewing of SAEs	Sponsor	Within 24 hours of receiving SAE form	Pharmacovigilance Committee
Reporting of SUSARs to MHRA	Sponsor	Within 7 days if life threatening or fatal. Within 15 days for others	Senior Research Governance Manager or delegate

16.6. Appendix 6 – Amendment History

Amendment No.	Protocol version no.	Author(s) of changes	Details of changes made
CTA review	2	Fiona McLaren-Neil, James Chalmers	Section 2 Rationale Arm 2: Disulfiram. Section 6.3.1 Exclusion criteria Arm 2: Disulfiram
AM02	3	Fiona McLaren-Neil, James Chalmers	Section 7.7.1. clarifications of sub-study procedures
AM03	4	Margaret band, James Chalmers	Addition of research bloods and sputum at visit 1, clarification of exacerbation definition.