Clinical Study Protocol

Drug Substance AZD9668

Study Code D0520C00002

Date [redacted]

A 2-week, randomised, double-blind, placebo-controlled, parallel group study to assess the tolerability and pharmacokinetics of orally administered AZD9668 in patients with COPD

Sponsor:

AstraZeneca AB, [redacted]

The following Amendment(s) and Administrative Changes have been made to this protocol since the date of preparation:

Amendment No.	Date of Amendment	Local Amendment No:	Date of Local Amendment

Administrative Change No.	Date of Administrative Change	Local Administrative Change No.	Date of Local Administrative Change

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PROTOCOL SYNOPSIS

A 2-week, randomised, double-blind, placebo-controlled, parallel group study to assess the tolerability and pharmacokinetics of orally administered AZD9668 in patients with COPD

Investigator

[redacted], physician

Study centre and number of patients planned

[redacted] It is planned to randomise 18 patients in this study.

Study period		**Phase of development**
Estimated date of first patient enrolled	[redacted]	II
Estimated date of last patient completed)	[redacted]	

The end of study is defined as the date of database lock, which is the time point after which no patient will be exposed to study-related activities.

Objectives

The primary objectives are:

1. To investigate the tolerability of 14 days' dosing with AZD9668 in patients with COPD, as measured by vital signs, ECG, lung function, haematology, clinical chemistry, urinalysis and adverse event reporting.

2. To determine the plasma pharmacokinetics and renal clearance of AZD9668 in patients with COPD.

The secondary objectives are:

1. To assess the effect of orally administered AZD9668 on neutrophils in induced sputum of patients with COPD.

2. To investigate the effects of AZD9668 on sputum bacteriology.

3. To measure the concentration of AZD9668 in induced sputum following oral administration.

The exploratory objectives are

1. To investigate the effect of AZD9668 compared to placebo treatment on cells and inflammatory markers in blood (including but not limited to TNFα, IL-8, serum amyloid A and high sensitivity C-reactive protein (hsCRP) in serum). These exploratory biomarkers may be reported separately, not as part of the main Clinical study report.

2. To investigate the clinical effects of AZD9668 by using the BronkoTest© diary card.

3. To investigate the effect of AZD9668 on markers of tissue degradation (including but not limited to urine and plasma desmosine, and sputum hydroxyproline). These exploratory biomarkers may be reported separately, not as part of the main Clinical study report.

4. To collect samples for possible retrospective pharmacogenetics analysis to investigate the influence of genotype on exposure (ADME), pharmacodynamic response including inter-individual biomarker response variability, tolerability and safety where appropriate. The results of the pharmacogenetic tests may be reported separately, not as part of the main Clinical study report.

Study design

This study is a randomised, double-blind, placebo-controlled, parallel-group, single-centre study to evaluate the tolerability and pharmacokinetics of AZD9668 administered for 14 days, in patients with moderate to severe COPD (GOLD stages II and III). 18 patients will be randomly assigned to receive either AZD9668 or matching placebo orally (in a ratio of 2 active: 1 placebo).

The pharmacogenetic research aspects of the study are not mandatory for the individual patients. Ethical approval is requested for these research components, in addition to, and separate from, approval for the main study.

Target patient population

Male and postmenopausal or surgically sterile female patients with moderate to severe COPD (GOLD stages II and III), aged between 40 to 80 years. Patients will be smokers or ex-smokers, with a history of >10 pack-years, post-bronchodilator FEV_1 30–80% predicted and FEV_1/FVC ratio < 70%.

Investigational product, dosage and mode of administration

AZD9668 will be administered as 30 mg tablets. On Day 1 patients will receive a single morning dose of 60 mg of AZD9668 (or matching placebo) for single dose PK. This will be

followed by daily doses of 60 mg b.i.d (twice daily, 12 hours apart) on Days 2 to 13 and finally a single morning dose of 60 mg on Day 14 for steady state PK.

Comparator, dosage and mode of administration

Matching placebo tablets will be given according to the same regimen as the IMP.

Duration of treatment

14 days.

Outcome variables

- **Primary outcome variables**
  - **Tolerability:**

    Adverse events, vital signs, ECG, lung functions, haematology, clinical chemistry, and urinalysis.
  - **Pharmacokinetic:**
    - AZD9668 plasma and urine concentrations
    - Day 1: C_{max}, t_{max}, $AUC_{(0-t)}$, $AUC_{(0-24)}$, C_{24}, AUC, $t_{1/2}$, CL/F, V_z/F, CL_R, A_e and F_e (%)
    - Day 7: $C_{min,ss}$
    - Day 14: $C_{min,ss}$, $C_{,max,ss}$, t_{max}, $AUC_{(0-t),ss}$, $AUC_{(0-24),ss}$, $C_{24,ss}$, AUC_{ss}, $t_{1/2ss}$, CL/F_{ss}, $V_z/F_{ss,}$ R_{ac}, CL_R, $A_{e,ss}$ and $F_{e,ss}$ (%)
- **Secondary outcome variables:**
  - Sputum absolute and differential neutrophil count
  - AZD9668 sputum concentrations
  - Quantitative sputum bacteriology
- **Exploratory variables**
  - Inflammatory markers in plasma (TNFα, IL-8, serum amyloid A, hsCRP)
  - Markers of tissue degradation (urine and plasma desmosine, sputum hydroxyproline)
  - BronkoTest© diary card data.

Statistical methods

Descriptive statistics will be used for all parameters in the study.

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LIST OF ABBREVIATIONS AND DEFINITION OF TERMS

The following abbreviations and special terms are used in this study protocol.

Abbreviation or special term	Explanation
α1-AT	Alpha 1-antitrypsin
ADME	Absorption, distribution, metabolism, excretion
AE	Adverse event (see definition in Section 4.7.1.1)
A_e	Cumulative amount unchanged drug excreted into urine
$A_{e,ss}$	A_e at steady state
ALT	Alanine aminotransferase (= GPT, SGPT)
AST	Aspartate aminotransferase (= GOT, SGOT)
AUC	Area under the plasma concentration-time curve from time zero to infinitive
$AUC_{(0-24)}$	AUC from time zero to 24 hours post-dose
$AUC_{(0-t)}$	AUC from time zero to time t.
$AUC_{(0-t),ss}$	$AUC_{(0-t)}$ at steady state
AUC_{ss}	AUC at steady state
$AUC_{(0-24),ss}$	$AUC_{(0-24)}$ at steady state
AZ	Astra Zeneca
BAL	Bronchoalveolar lavage
BMI	Body mass index
C_{24}	Concentration at 24 hours post-dose
$C_{24,ss}$	C_{24} at steady state
CL/F	Apparent plasma clearance following oral drug administration
$CL/F_{,ss}$	CL/F at steady state
CL_R	Renal clearance of drug from plasma
C_{max}	Observed peak or maximum plasma concentration following drug administration
$C_{max,ss}$	C_{max} at steady state
$C_{min,ss}$	Minimum plasma (trough) drug concentration after repetitive dosing after steady state is achieved, prior to the first dose of the study day
COPD	Chronic obstructive pulmonary disease
[redacted]	[redacted]
CPK	Creatinine phosphokinase

Abbreviation or special term	Explanation
CPU	Clinical pharmacological unit
CRF	Case Report Form
CS	Cardiosoft®
CSA	Clinical Study Agreement
CSR	Clinical study report
DMPK	Drug metabolism and pharmacokinetics
DNA	Deoxyribonucleic acid
ECG	Electrocardiogram
eCRF	electronic CRF
EDTA	Ethylenediaminetetraacetic acid
eSource	Electronic source
Ethics Committee	Synonymous to Institutional Review Board and Independent Ethics Committee
EU	European Union
F_e (%)	Percentage of the dose excreted as unchanged parent in the urine
$F_{e,ss}$ (%)	F_e(%) at steady state
FEV_1	Forced expiratory volume in the first second
FSH	Follicle-stimulating hormone
FTiM	First-trial-in-man
FVC	Forced vital capacity
GCP	Good Clinical Practice
GEMS	General Electrics Medical Systems
GGT	Gamma glutamyltransferase
GMP	Good Manufacturing Practice
GOLD	Global Initiative for Chronic Obstructive Lung Disease
HDPE	High-density polyethylene
HELC	Human Exposure Limits Committee
hERG IC50	Human Ether A-Go-Go Related Gene
HIV	Human immunodeficiency virus
hsCRP	High sensitivity C-reactive protein
IC	Inspiratory capacity
ICH	International Conference on Harmonisation

Abbreviation or special term	Explanation
ICS	Inhaled corticosteroids
IL1-ß	Interleukin 1 beta
IL-8	Interleukin-8
IMP	Investigational medicinal product
International Co-ordinating investigator	If a study is conducted in several countries the International Co-ordinating Investigator is the Investigator co-ordinating the investigators and/or activities internationally.
IPS	Investigational Product Supplies
ISF	Investigator Site File
LABA	Long-acting beta agonist
LAMA	Long-acting muscarinic antagonist
LC-MS/MS	Liquid chromatography with tandem mass spectrometry
LDH	Lactate dehydrogenase
LFT	Liver function test
LIMS	Laboratory Information Management System
LLN	Lower limit of normal
LLOQ	Lower limit of quantification
LTB4	Leukotriene B4
MAD	Multiple ascending dose
MPO	Myeloperoxidase
MTD	Maximum tolerated dose
NB	Lat. Nota Bene
NE	Neutrophil elastase
NOAEL	No observable adverse effect level
NOEL	No observed effect level
NSAIDS	Non-steroidal-anti-inflammatory drugs
OAE	Other Significant Adverse Event (ie, adverse events of particular clinical importance, other than SAEs and those AEs leading to discontinuation of the patient from study treatment; see definition in Section 4.7.1.1).
pCRF	Paper CRF
PK	Pharmacokinetic(s)
PRO(s)	Patient-Reported Outcomes

Abbreviation or special term	Explanation
QT	ECG interval measured from the beginning of the Q wave (or the R wave if Q is missing) to the end of the T wave; the time interval of ventricular depolarisation and repolarisation
QTc	Corrected QT
R_{ac}	Accumulation ratio
RBC	Red blood count
SABA	Short-acting beta agonist
SAD	Single ascending dose
SAE	Serious adverse event (see definition in Section 4.7.1.1).
SAMA	Short-acting muscarinic antagonist
SAP	Statistical analysis plan
SRC	Safety Review Committee
SVC	Slow vital capacity
$t_{1/2}$	Terminal half-life of drug in plasma
$t_{1/2,ss}$	$t_{1/2}$ at steady state
t_{max}	Time to reach observed peak or maximum concentration following oral drug administration
$t_{max,ss}$	t_{max} at steady state
UK	United Kingdom
ULN	Upper limit of normal
V_z/F	Apparent terminal volume of distribution following extra-vascular dosing
$V_z/F_{,ss}$	V_z/F at steady state
WHO	World Health Organisation

1. INTRODUCTION

1.1 Background

Chronic obstructive pulmonary disease (COPD) is the fourth leading cause of morbidity and mortality in the US (US Department of Health and Human Services 1998) and worldwide is projected to rank fifth in burden of disease in 2020 (Lopez 1998). The prevalence of physiologically defined COPD in adults, aged 40 years or over, is around 9-10% (Halbert 2006). A significant proportion of these patients suffer from severe disability, and this subset of patients contributes largely to the health care costs of COPD (Rutten van Molken 1999).

COPD is a chronic lung disease, characterized by airflow limitation that is not fully reversible, and this is associated with an abnormal chronic inflammatory response in the respiratory system to noxious particles or gases (GOLD 2005). Currently, the mainstay of treatment of airflow limitation is bronchodilators. Despite a prominent inflammatory response in the airways in COPD, in contrast to asthma, anti-inflammatory therapy with corticosteroids is relatively ineffective. Therefore, there is a need to explore the possibilities of developing other therapies for COPD.

Neutrophil elastase (NE) is a serine protease found in high levels in neutrophils. NE is able to degrade extracellular matrix and proteins leading to long-term destruction of the lung parenchyma. Inhibition of neutrophil elastase has the potential to inhibit this proteolytic destruction raising the possibility of disease modification. If, as predicted from in vivo studies, NE inhibition can affect not only lung destruction but also epithelial metaplasia, goblet cell hyperplasia and the loss of cilia caused by exposure to cigarette smoke, as well as infiltration of inflammatory cells, shorter-term symptomatic benefits are also possible with this approach.

There are a number of clinical and experimental observations supporting a role for NE in COPD. In α1-AT deficiency, an established genetic risk factor, the development of emphysema is believed to be caused by the unchecked action of proteases on lung tissue. Cigarette smoking, a major cause of COPD, induces a functional antiprotease deficiency in the lower respiratory tract of humans. There is a strong relationship between the severity of emphysematous change in human lung and the amount of NE in lung interstitium. NE in bronchoalveolar lavage (BAL) fluid correlates directly, and anti-elastase capacity inversely, with emphysema severity. Desmosine excretion, a biological marker of lung destruction, is significantly increased in patients with COPD as compared to healthy smokers.

Airway inflammation plays an important role in the pathogenesis of COPD. Neutrophils are recognized as major cellular mediators of inflammation and play a central role in many of the features of COPD. Sputum from COPD patients has significantly greater numbers of neutrophils and the neutrophil specific marker myeloperoxidase (MPO) in sputum correlates with the NE concentrations. The activation of neutrophils results in the release of LTB4 and IL-8, which further contributes to a self-perpetuating inflammatory response in COPD.

In the guinea pig, the NE inhibitor AZD0892 inhibited lung neutrophilic inflammation and emphysema development due to chronic inhalation of cigarette smoke. Data from experimental studies suggest that NE knock out mice are partially (59%) protected against development of emphysema in the smoking model. Transgenic mice that express extremely low levels of α1-AT were partially (63%) protected against emphysema development in a 6 month smoking study when treated with human α1-AT. Studies in rat models have shown that oral administration of neutrophil elastase inhibitor ONO-6818, attenuated lung haemorrhage and accumulation of neutrophils in the lung induced by human NE, and long-term administration of ONO-6818 prevented human NE-induced lung emphysematous changes. In addition, there are a number of publications supporting a key role for NE in experimental models of mucus hypersecretion of COPD.

AZD9668, a pyridone of molecular weight of 545.5 g/mol, is a potent, oral, selective, reversible inhibitor of human NE. It is being developed as a possible therapeutic agent both for symptomatic treatment as well as disease modification in COPD. Orally administered AZD9668 produced a dose-dependent inhibition of human NE-induced lung haemorrhage in the mouse. Mice exposed to cigarette smoke and treated with AZD9668 showed a significant reduction in total number of cells and neutrophils and IL1-β, compared to vehicle treated smoke exposed mice, indicating the possibility of inhibiting endogenously released NE with this drug.

AZD9668 was well tolerated in rat and dog toxicity studies. In a dog 28-day toxicity study minor hematological abnormalities were noted at high doses. There was no bone-marrow pathology. Electrophysiological studies showed a hERG IC50 of 69 μM, and *in vivo* a slight QT-prolongation (9%) at a high dose in a dog telemetry study. Mutagenicity tests indicated that AZD9668 does not pose a significant genotoxicity risk.

A first administration to man, Phase I, randomised, double blind, placebo-controlled, 2-part study (D0520C00001) to assess the safety and tolerability and pharmacokinetics of single and multiple oral doses of AZD9668 in healthy volunteers has been completed.

In the single ascending dose part of the study, AZD9668 was administered at doses of 2 mg, 10 mg, 30 mg, 60 mg, 120 mg and 150 mg with 36 subjects receiving the active drug and 12 subjects receiving the placebo. No serious adverse events occurred. Fifty-seven non-serious adverse events were reported by 27 subjects. The AEs were generally of the type commonly observed in Phase-I studies. The severity for the majority of the events was mild; 6 were reported as moderate, and for one subject 2 events were reported as severe. The AEs considered to be moderate were headache (2 cases – 30 mg & 150 mg), migraine (2 cases, 120mg & placebo) and influenza-like illness (1 case, 30mg) and dizziness (1 case, 120mg). The AEs considered to be severe were a syncope associated with a bradycardia, 4 hours 36 minutes after a 2 mg dose (this event resolved with supportive care within 3 minutes) The most frequently reported adverse event was headache (14/48 subjects). Five subjects reported headache on the 150mg dose (mild in 4 and moderate in 1). In comparison only 3 out of 12 subjects on placebo reported headache. In the subject who developed a moderate headache on the 150 mg dose, the Investigator considered this causally related to the study drug. One

subject on the 120 mg dose developed a non-sustained broad complex monomorphic tachycardia (5 beats, 2 seconds) 37 minutes after dosing. This subject was noted to have frequent ventricular ectopics prior to dosing. The investigator considered this event not related to the study drug. No laboratory abnormalities of any concern or QT prolongation were noted in any of the subjects.

In the multiple ascending dose part of the study the drug was given in doses of 30 mg, 70 mg and 120 mg once daily for 8 days. Twenty-seven subjects were randomised; 9 to placebo, and 6 to each of the dosing groups. No serious adverse events were reported; 47 non-serious adverse events were reported by 20 subjects. Again the majority of AEs were mild, 2 were moderate and one severe. The AEs considered to be moderate were headache (on 30 mg dose) and dyspepsia (on placebo). Severe AE was a headache on the 120 mg dose. Three subjects were withdrawn; one on the 30 mg dose because of asymptomatic nocturnal bradycardia, one on the 70 mg dose because of a run of premature atrial contractions, and one on the 120 mg dose because of headache, nausea and vomiting. The AEs on the 120 mg dose were deemed to be causally related to study drug. No significant laboratory or QT abnormalities were noted in this part of the study.

Overall, in this first administration to man study, AZD9668 was well tolerated except for the occurrence of headaches at the highest doses tested. The PK data from this study is detailed in Section 3.2.1.

Further details of the preclinical toxicology and pharmacology can be found in the Investigators Brochure.

1.2 Rationale

AZD9668 is primarily being developed as a treatment for COPD. So far it has been found to be safe and tolerated in healthy volunteers. Before embarking on long-term proof-of-concept studies in subjects with COPD, it is necessary to establish the safety and tolerability of AZD9668 in this target patient population. It is also necessary to establish the PK of the drug in COPD patients. These are the main purposes of the study.

2. STUDY OBJECTIVES

2.1 Primary objective

The primary objectives of this study are

1. To investigate the tolerability of 14 days' dosing with AZD9668 in patients with COPD, as measured by vital signs, ECG, lung function, haematology, clinical chemistry, urinalysis and adverse event reporting.

2. To determine the plasma pharmacokinetics and renal clearance of AZD9668 in patients with COPD.

2.2 Secondary objectives

1. To assess the effect of orally administered AZD9668 on neutrophils in induced sputum of patients with COPD.

2. To investigate the effects of AZD9668 on sputum bacteriology.

3. To measure the concentration of AZD9668 in induced sputum following oral administration.

2.3 Exploratory objectives

1. To investigate the effect of AZD9668 compared to placebo treatment on cells and inflammatory markers in blood (including but not limited to TNFα, IL-8, serum amyloid A and high sensitivity C-reactive protein (hsCRP) in serum).

2. To investigate the clinical effects of AZD9668 by using the BronkoTest© diary card.

3. To investigate the effect of AZD9668 on markers of tissue degradation (including but not limited to urine and plasma desmosine, and sputum hydroxyproline.

4. To collect samples for possible retrospective pharmacogenetics analysis to investigate the influence of genotype on exposure (ADME), pharmacodynamic response including inter-individual biomarker response variability, tolerability and safety where appropriate. The results of the pharmacogenetic tests may be reported separately, not as part of the Clinical study report of this study.

3. STUDY PLAN AND PROCEDURES

3.1 Overall study design and flow chart

This Clinical Study Protocol has been subjected to a peer review according to AstraZeneca standard procedures.

This study is a randomised, double-blind, placebo-controlled, parallel-group, single-centre study to evaluate the tolerability and pharmacokinetics of AZD9668 administered for 14 days in patients with moderate to severe COPD (GOLD stages II and III). 18 patients will be randomly assigned to receive either AZD9668 or matching placebo orally (in a ratio of 2 active: 1 placebo). On Day 1 patients will receive a single morning dose of 60 mg of AZD9668 (or matching placebo) for single dose PK. This will be followed by daily doses of 60 mg bid (twice daily, 12 hours apart) on Days 2 to 13 and finally a single morning dose of 60 mg on Day 14 for steady state PK.

The study will consist of 5 visits.

Patients must provide full written informed consent before any study-related procedures, restrictions or screening assessments are carried out.

Visit 1: Screening visit (Day –21 to Day –8): At Visit 1, after having given written informed consent, patients will be enrolled into the study and screened for eligibility (Table 1). If eligibility is confirmed at Visit 1, patients will be issued with a BronkoTest© diary card (to be completed daily for the remainder of the study) and asked to return to the CPU at Visit 2.

Visit 2: Treatment Period (Day –1 to Day 3): The day before dosing is defined as Day –1 and the day of dosing as Day 1, Day 0 does not exist.

Patients will attend the CPU in fasted state with their BronkoTest© diary card on the morning of Day –1 and stay until Day 3. Eligibility will be re-confirmed and safety assessments performed (vital signs, ECG, lung function, haematology, clinical chemistry and urinalysis, urine drug screen including alcohol). Twenty-four hour ambulatory cardiac monitoring (telemetry) will be started on Day –1 and monitoring continued for 24 hours post first dose. All urine produced over the 24 hours before the first dose will be collected for assessment of baseline desmosine levels and for the pre-dose urine PK. [For all timed urine collections, the patients should empty the bladder within 10 minutes of Time 0, discard that urine and then collect all the urine from then onwards. At Time (0 + t hours) they should then empty the bladder and add that to the collection.] On the morning of Day –1, the patients will provide an induced sputum sample; weight of the sample will be recorded, and the sample processed for neutrophils and AZD9668 levels. Blood will also be collected for baseline assessment of hsCRP and serum amyloid A.

Patients will fast from midnight on Day 1 until 2 hours after the first dose (water will be allowed for thirst up until 1 hour before dosing).

On Day 1, pre-dose patients will be asked to provide a spontaneous sputum sample for quantitative bacteriology. They will be randomised to receive either AZD9668 or placebo: A single dose will be administered in the morning of Day 1. All urine produced over the 24 hours after the first dose will be collected for measurement of AZD9668 concentrations. Urine will be collected for time periods 0-4 hours, 4-12 hours and 12-24 hours. Aliquots from each of these collections will be taken for urine PK, and an aliquot from the pooled sample will be taken for urine desmosine. Serial blood samples will be collected for PK analysis at pre-dose and at 0.25, 0.5, 1, 2, 3, 4, 5, 6, 8, 12, and 24 hours post dose (NB. PK samples taken at 24 h will be taken pre-dose). Vital signs and ECG will be done at approximately 1 hour after the first dose.

On Days 2 - 13 the patients will receive 2 daily doses of AZD9668. The first dose of each day will be taken at approximately the same time (± 1h) as the first dose administered on Day 1 and the 2nd daily dose will be approximately 12 hours later (± 1 hour).

Further safety assessments (vital signs, ECG, lung function, haematology, clinical chemistry and urinalysis) will be done before the morning dose on Day 2. Patients will be discharged on the morning of Day 3 (after having taken the 4th dose of the study drug). Prior to discharge,

patients will be re-issued with their BronkoTest© diary card and sufficient tablets to allow daily self-dosing until their next visit. Vital signs will be measured and an ECG recorded before discharge. The pharmacogenetic sample can be taken at any convenient time point after the first dose.

The BronkoTest® diary card will be filled in for all days whilst the subject is in the CPU and at home.

Visit 3: Treatment period (Day 7 ±1 day): Patients will attend the CPU on the morning of Day 7 (±1 day) in fasted state, having fasted from midnight (water will be allowed for thirst up until 1 hour before dosing) and having withheld taking their morning dose of study medication for that day. Patients will bring with them their BronkoTest© diary card and any remaining study medication. Before administration of the morning dose, safety assessments (vital signs, ECG, lung function, haematology, clinical chemistry, urinalysis and urine drug screen) will be performed. Blood samples will be collected pre-dose for PK and for assessment of hsCRP and serum amyloid A. Patients will then take their morning dose of study medication in the CPU. Patients will be discharged from the CPU with their BronkoTest© diary card and sufficient tablets to allow daily self-dosing until their next visit.

Visit 4: End of Treatment (Days 14 & 15 ±1 day): Patients will attend the CPU with their BronkoTest© diary card and study medication on the morning of Day 14 in fasted state, having fasted from midnight (water will be allowed for thirst up until 1 hour before dosing) and having withheld taking their morning dose of study medication, and stay until the morning of Day 15. Blood will also be collected pre-dose for assessment of PK, hsCRP and serum amyloid A. Before administration of the morning dose on Day 14, safety assessments (vital signs, ECG, lung function, haematology, clinical chemistry, urinalysis and urine drug screen) will be performed. Patients will then take their morning dose of study medication in the CPU. This will be the final dose of study medication. At 1 hour post-dose, an ECG will be recorded. Approximately 1-2 hours after this last dose, patients will provide an induced sputum sample (around the same time they had the induced sample on Day –1); weight of the sample will be recorded, and the sample processed for neutrophils and AZD9668 levels.

All urine produced over the 24 hours after the last dose will be collected for measurement of desmosine levels and AZD9668 concentrations. Urine will be collected for time periods 0-4 hours, 4-12 hours and 12-24 hours. Aliquots from each of these collections will be taken for urine PK, and an aliquot from the pooled sample will be taken for urine desmosine. Serial blood samples will be collected for PK analysis at pre-dose and at 0.25, 0.5, 1, 2, 3, 4, 5, 6, 8, 12, and 24 hours post dose. On the morning of Day 15, a spontaneous sputum sample will be collected for quantitative bacteriology, and the patients will be discharged from the CPU with their BronkoTest© diary card to bring to their follow-up visit.

Visit 5: Follow-up (Day 21 ±3 days): Patients will return to the CPU with their BronkoTest© diary card in fasted state, having fasted from midnight (water allowed), for follow-up safety assessments (physical examination, vital signs, ECG, lung function, haematology, clinical

chemistry and urinalysis). A blood sample for hsCRP and serum amyloid A will also be collected at this visit.

Throughout the study, lung function measurements should be performed at the same time of day ± 2 hours in relation to the time point at Visit 1 (baseline).

Each patient will attend the visits as outlined in Figure 1.

Figure 1 Study flow chart

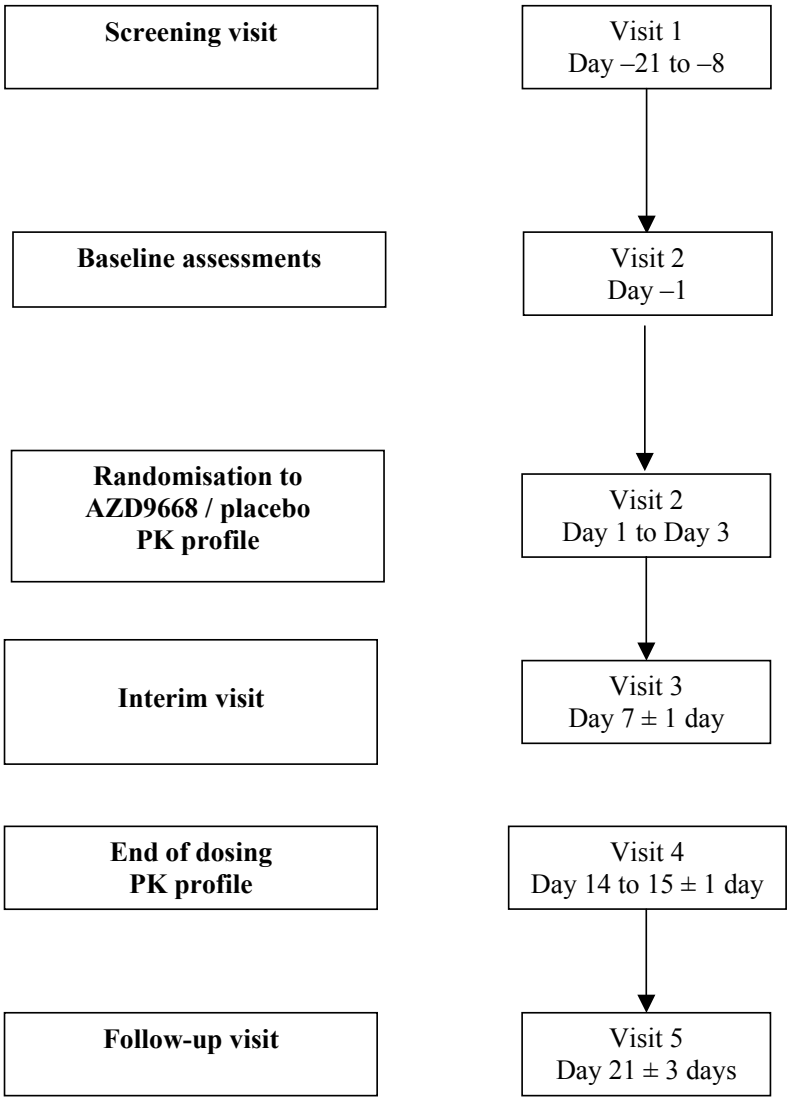

Table 1 Study plan

	Screening visit	Study Visit (start)				Study visit	Study visit (end)		Follow-up
Study Visit	**1**	**2**				**3**	**4**		**5**
Study Day	**-21 to -8**	**-1**	**1**	**2**	**3**	**7 (±1)**	**14(±1)**	**15**	**21 (±3)**
Residential stay [a]		✓	✓	✓	✓		✓	✓	
Informed consent	✓								
Incl./Excl criteria	✓	✓							
Demographics	✓								
Med/surgical history	✓								
Drug screen including alcohol	✓	✓				✓	✓		
Virology	✓								
Pregnancy test /FSH (if appropriate)	✓								
Concomitant medication	✓	✓	✓	✓	✓	✓	✓	✓	✓
Adverse event questioning		✓	✓	✓	✓	✓	✓	✓	✓
Randomisation			✓						
Dispense study drug					✓	✓			
Physical examination	✓								✓
Vital signs [b]	✓	✓	✓	✓	✓	✓	✓		✓
12-lead ECG [b]	✓	✓	✓	✓	✓	✓	✓		✓
Telemetry		✓	✓						
Safety blood/urinalysis [c]	✓	✓		✓		✓	✓		✓
Lung function [c]	✓	✓		✓		✓	✓		✓
Induced sputum [d]		✓					✓		
Quantitative sputum bacteriology[e]			✓					✓	
24-h urine collection for desmosine[f]		✓					✓		
Urine PK[g]		✓	✓				✓		
Plasma - full PK profile[h]			✓				✓		
Plasma– single PK sample[i]						✓			
Serum – hsCRP, amyloid-A		✓				✓	✓		✓
Plasma for multiplex analysis of cytokines (including but not limited to TNF-α, IL-8)		✓					✓		✓

Table 1 Study plan

	Screening visit	Study Visit (start)				Study visit	Study visit (end)		Follow-up
Study Visit	**1**	**2**				**3**	**4**		**5**
Study Day	**-21 to -8**	**-1**	**1**	**2**	**3**	**7 (±1)**	**14(±1)**	**15**	**21 (±3)**
BronkoTest© diary card[j]	✓	✓	✓	✓	✓	✓	✓	✓	✓
Return study drug						✓	✓		
Non-residential visit	✓					✓			✓
Genetic blood sample[k]			✓						

[a] Patients to be resident in the CPU from the morning of Day -1 to the morning of Day 3, and from the morning of Day 14 to the morning of Day 15.

[b] Vital signs & ECG to be taken at Screening visit, Day –1, Day 1 (1h after 1st morning dose), Days 2, 3 (prior to discharge), 7 and 14 (before morning dose), and at follow-up. An additional ECG will be recorded 1 hour post-dose in Day 14..

[c] Safety blood, urinalysis & lung function (SVC, FEV_1, FVC, IC) to be done at Screening visit, Day –1, before morning dose on Days 2, 7 & 14, and at follow-up.

[d] Induced sputum (for sputum weight, neutrophils, & AZD9668) to be done at visit 2 (Day –1)and 1-2 hours after the last dose on Day 14.

[e] Quantitative sputum bacteriology to be done on a spontaneous sample (pre-dose on Day1), preferably that produced first thing in the morning.

[f] Urine for desmosine will be collected during the following periods: 24 h period before the first dose (Days –1 to 1) and 24 h period after the last dose (Days 14-15). For the latter, when it coincides with the urine PK, the timed PK samples will be pooled to take an aliquot for urine desmosine.

[g] For urine PK aliquots will be taken from samples collected for the following periods: 0-4 hours, 4-12 hours and 12-24 hours. On Day –1 only, an aliquot will be taken from the 24-hour collection for the pre-dose sample.

[h] Serial PK plasma samples to be taken pre-dose and 0.25, 0.5, 1, 2, 3, 4, 5, 6, 8, 12, and 24 hours after the first and last dose (i.e. on Days 1-2 and 14-15). NB the sample taken at 24 hours on Day 2 will be taken pre-dose.

[i] Plasma PK sample taken pre-dose

[j] BronkoTest© diary card to be completed daily by patient throughout the entire study period.

[k] If consent is not obtained at Visit 1, it can be obtained at any of the subsequent visits. Sampling is planned for the randomisation visit, but can be taken at a subsequent visit

3.2 Rationale and risk/benefit assessment

3.2.1 Rationale for study design, doses and control groups

Patients with COPD are the target population for the NE inhibitor AZD9668. The main purpose of this study is to establish the tolerability of AZD9668 in patients with COPD as a preliminary to long-term proof-of-concept studies in this population. It is also intended to get pharmacokinetic (PK) data in this population that would help in the planning of future patient studies. So far safety/tolerability and PK data are available in healthy volunteers up to 150 mg as a single dose and up to 120 mg daily for 8 days as multiple doses. A 14-day study in 12

patients with COPD on active treatment is deemed sufficient to establish tolerability in a patient population, before proceeding to longer-term studies in patients with COPD.

The first-in- man, two-part, single and multiple ascending dose (SAD and MAD) study of AZD9668 in healthy volunteers (D0520C00001) is now complete and the reporting phase is underway. In the SAD part of the study the doses that have been safely administered were 2, 10, 30, 60, 120 and 150 mg. Dose escalation was stopped because the exposure limits set by the internal AstraZeneca Human Exposure Limits Committee (HELC) were reached. These limits (i.e. $AUC_{(0-24h)}$ and C_{max} of 18 µM.h and 2.5 µM, respectively) were set following a review of the data obtained in the 1-month toxicology studies in the rat and dog and have been based upon the exposures at the no observed adverse effect level (NOAEL) in rat (370 mg/kg), corrected for the differences in plasma protein binding between the species. The NOAEL in the rat has been used because the free exposures at this dose are lower than those obtained in the dog at the no observed effect level (NOEL) dose. In the MAD part of the study the doses that have been safely administered were 30, 70 and 120 mg once a day for a total of 8 days.

Following single dose administration of AZD9668 (2 - 150 mg) absorption was rapid, median t_{max} ranged from 1h to 2h. The geometric mean CL/F ranged from 15.4 to 21.6 L/h and the geometric mean V_z/F ranged from 157 to 334L. The geometric mean terminal half-life of AZD9668 ranged from 5.04 to 14.99h. The CL/F, V_z/F and $t_{1/2}$ appeared to be independent of dose. The exposure (C_{max} and AUC) increased proportionally with dose between doses of 10mg and 150mg.

AZD9668 is renally eliminated and renal clearance (CL_R) and the percentage of the dose excreted as unchanged AZD9668 in the urine over 48 h ($F_{e(0-48)}$ (%)) appeared to be independent of dose. The geometric mean CL_R ranged from 5.83 L/h to 6.98 L/h, and the range of the geometric mean $F_{e(0-48)}$ (%) was 31.6 % to 40.0 %.

Following multiple dose administration of AZD9668 (30 mg, 70 mg and 120 mg, once a day for 8 days), absorption was similar following single and multiple dosing; the median t_{max} and $t_{max,ss}$ was 1h for all dose groups. The geometric mean $CL/F_{,ss}$ was 14.7, 14.8 and 19.4 L/h and the geometric mean $Vz/F_{,ss}$ was 191L, 218 and 245L, respectively. The geometric mean $t1/2_{,ss}$ was 9.04h, 10.24h and 8.73h, respectively, and there was no evidence for time or dose dependent change in PK. There was no evidence of accumulation upon multiple dosing to steady state.

AZD9668 is renally eliminated and renal clearance at steady state ($CL_{R,ss}$) and the percentage of the dose excreted as unchanged AZD9668 in the urine at steady state over 48 h ($F_{e(0-48),ss}$ (% dose)) appeared to be independent of dose. The geometric mean $CL_{R,ss}$ was 5.29 L/h, 5.70 L/h and 5.83 L/h, and the geometric mean $F_{e(0-48),ss}$ (% dose) was 38.8 %, 40.9 % and 31.6 %, respectively.

Administration of AZD9668 after consumption of a high fat meal resulted in a reduction in the rate of absorption (median t_{max} was 4 h, 3 h and 4 h for 30 mg, 70 mg and 120 mg, respectively). The C_{max} was reduced by 22 %, 31 % and 22 % and the $AUC_{(0-24h)}$ was slightly

reduced, by 8 %, 5 % and 5% for 30 mg, 70 mg and 120 mg, respectively. Thus consumption of the food reduced the rate but had relatively little effect on the overall extent of absorption.

A population PK model of the PK data from the SAD and MAD study has been developed and was used to estimate the mean population PK parameters of AZD9668 with the associated variability and to assess the affect of consumption of a high fat breakfast on the PK of AZD9668. A dose of 60 mg twice a day has been selected as the dose for the current study. This dose has been chosen as a dose that is predicted to achieve trough steady state therapeutic plasma concentrations of 19 nM (predicted from an in vivo mouse acute lung injury model) in all patients, whilst ensuring that the predicted geometric mean $C_{max,ss}$ and $AUC_{(0-24h),ss}$ are below the HELC limits. The estimated margins to the HELC limits in the fasted state are 2.6 and 1.4 for $C_{max,ss}$ and $AUC_{(0-24h),ss}$, respectively, and this will be the dose that will be used in the subsequent proof-of-principle studies with AZD9668. If a dose of 60 mg b.i.d is not tolerated by an individual patient then his/her dose may be halved to 30 mg b.i.d for the remainder of the study, at the investigator's discretion. The population PK model predicts that this will still achieve trough steady state plasma concentrations above 19 nM in all the patients. Since the objective of the study is to investigate tolerability, the higher dose (i.e. 60 mg b.i.d) has been chosen as the study dose rather than the minimum dose that is predicted to achieve therapeutic exposures. AZD9668 will only be dosed once a day on Days 1 and 14 to enable measurement of single dose and steady state pharmacokinetics respectively.

From the population PK modelling it was identified that the bioavailability of AZD9668 was approximately 90% of that when it was administered in the fasted state. This did not however have any impact upon the dose selection. For the subsequent studies therefore, it is not necessary for the patients to restrict food around administration of their doses.

Patients will receive their morning dose in a fasted state on Days 1, 7 and 14, i.e. on days when they are having ECG measurements. This ensures that the ECG measurements can be taken when C_{max} is maximised.

3.2.2 Risk/benefit and ethical assessment

The patients will have no individual benefit from participation in the study. The early development studies are to enable further investigations to evaluate AZD9668 as a treatment of COPD.

In the FTiM single and multiple dose-escalation study of AZD9668, the AEs that were reported included headache, nausea and vomiting. Some ECG abnormalities were also noted during 24-hour ambulatory monitoring, which included nocturnal bradycardia, a short run of premature atrial contractions and a non-sustained broad-complex tachycardia. Also one subject experienced syncope associated with a bradycardia. With the exception of headaches, which occurred consistently at high doses, there was no reason to suspect that any of the other AEs could be related to AZD9668, as no consistent pattern was discernible and there was no clear dose-response relationship. There was no QT prolongation noted at any of the doses. Nevertheless, patients with a QT > 450 msec will be excluded from this study, as pre-clinical toxicology showed some tendency to QT prolongation in dog telemetry studies, albeit at high

doses. The ECG abnormalities noted on 24-hour ambulatory monitoring seemed to occur in subjects who already had some similar baseline abnormalities. For this reason, patients with any baseline arrhythmia, including premature ventricular and supraventricular ectopics exceeding 15/min, will be excluded from this study.

No significant laboratory abnormalities of any significance were observed in the FTiM study. However, the haematological and biochemical abnormalities noted in pre-clinical toxicological studies will continue to be monitored in this study also. In addition, patients with a calculated creatinine clearance of <70 ml/min will be excluded as the PK from the FTiM study indicates that the drug is renally eliminated. Excluding these patients would mitigate the risk of high drug exposures related to renal impairment.

For an overall risk benefit/ethical assessment of developing a NE inhibitor in COPD, see the Investigator's Brochure.

3.3 Selection of study population

3.3.1 Study selection record

Investigators must keep a record of patients who were considered for enrolment but were never enrolled e.g. patient screening log. This information is necessary to establish that the patient population was selected without bias.

3.3.2 Inclusion criteria

For inclusion in the study treatment period patients must fulfil all of the following criteria:

1. Be willing and able to comply with study procedures and provide written informed consent

2. Be male or post-menopausal (for more than 12 months)/surgically sterile female between 40 and 80 years

3. Have a clinical diagnosis of COPD

4. Smokers or ex-smokers (with at least 10 pack years)

5. Post-bronchodilator FEV_1 30 - 80% predicted, FEV_1/FVC ratio < 70%

For inclusion in this genetic research, patients must fulfil the following criterion:

1. Provision of informed consent for genetic research

If a patient declines to participate in the genetic research, there will be no penalty or loss of benefit to the patient. The patient will not be excluded from other aspects of the study described in this Clinical Study Protocol, providing consent has been obtained.

3.3.3 Exclusion criteria

Any of the following is regarded as a criterion for exclusion from the study:

1. Concomitant diagnosis of significant pulmonary disease other than COPD, including symptomatic asthma, cystic fibrosis and allergic bronchopulmonary aspergillosis

2. An acute exacerbation (defined as an increase in respiratory symptoms requiring hospitalisation and/or a course of oral glucocorticosteroids and/or antibiotics, either prescribed or self administered); or acute respiratory infection (upper or lower) in the 4 weeks prior to Visit 1 or Visit 2

3. Use of oral corticosteroids in the 8 weeks prior to Visit 2 (use of inhaled corticosteroids is allowed)

4. Use of antibiotics, systemically or nebulised, in the 4 weeks prior to Visit 1 or Visit 2

5. Current requirement for oxygen therapy

6. Any other clinical disease or disorder (including insulin-dependent diabetes) which, in the opinion of the investigator, may either put the patient at risk because of participation in the study, or may influence the results of the study, or the patient's ability to participate in the study

7. Positive test result for HIV, Hepatitis B or C

8. Scheduled in-patient surgery or hospitalisation during the course of the study

9. Previous randomisation into the present study

10. Participation in another clinical study involving an investigational product within 4 weeks or 5 times the half life, whichever is longer, of Visit 1

11. A past history of or current clinical or laboratory evidence of renal disease, or a calculated creatinine clearance (Cockcroft-Gault formula) of ≤70 ml/min at Visit 1

12. QT_c > 450 msec or the presence of any arrhythmia in the ECG at Visit 1. (Premature ventricular or supraventricular ectopics up to 15/minute will be allowed as long as there are no other associated cardiac abnormalities).

13. Any clinically relevant abnormal findings in physical examination, clinical chemistry, haematology, urinalysis, vital signs, ECG or lung function at Visit 1, which, in the opinion of the investigator, may either put the patient at risk because of participation in the study, or may influence the results of the study, or the patient's ability to participate in the study.

14. Patients who, in the opinion of the investigator, should not participate in the study.

15. History of excessive alcohol consumption or chronic alcohol induced disease

16. If participation in the study would result in the volunteer donating more than 1350 mL of blood in the 12 months or 500 mL of blood in the 3 months before the end of the study

17. The patient is not able to understand and comply with protocol requirements, instructions and protocol-stated restriction

18. Vulnerable patients (e.g. persons kept in detention).

19. Involvement in the planning and conduct of the study (applies to both AstraZeneca staff or staff at the study site)

3.3.4 Restrictions

1. Patients should not use oral steroids or change their inhaled corticosteroids or dosage throughout the study, unless directed by the investigator

2. Patients should withhold the use of short acting bronchodilators for 4 hours, and long acting bronchodilators for 12 hours prior to lung function assessments.

3. Use of disallowed concomitant medication (refer to Section 3.7).

4. Patients should not donate blood at any time during the study or for 3 months following completion of the study

5. Fast from the midnight (24:00) prior to dosing until 2 hours post-dose on PK sampling days, i.e. Days 1, 7 ±1 and 14. Water is allowed up to 1 hour before drug administration and will be allowed from 2 hours after drug administration

6. Abstain from alcohol for 72 hours prior to blood and urine samples are taken at the screening and post study medical visits. Patients will also be required to abstain from alcohol for 72 hours prior to each visit in the CPRU during each study visit.

7. Abstain from taking part in any other study, whilst participating in this study.

8. Male patients should abstain from unprotected sex and sperm donation from date of consent until 4 months after last dosing. Recommended contraception will be double barrier method, i.e. condom and in addition, the female partner to use a reliable contraceptive method, or they must refrain from sexual intercourse during the entire clinical study.

9. There is no evidence from clinical experience to date that AZD9668 is capable of inducing photosensitivity. However until further studies are concluded, patients

should be advised not to sunbathe or use sunbeds or U-V lamps, and to limit exposure to sunlight as much as practicable during dosing and for 1 week after last dose of study medication.

3.3.5 Discontinuation of patients from treatment or assessment

3.3.5.1 Criteria for discontinuation

Patients may be discontinued from study treatment and assessments at any time at the discretion of the Investigator. The following could be specific reasons for discontinuing a patient from the study.

- Voluntary discontinuation by the patient who is at any time free to discontinue his/her participation in the study, without prejudice to further treatment
- Safety reasons as judged by the investigator and/or AstraZeneca
- Severe non-compliance to protocol as judged by the investigator and/or AstraZeneca
- Incorrect enrolment, i.e. the patient does not meet the required inclusion/exclusion criteria for the study
- Patient lost to follow-up
- Disease exacerbation, defined as an increase in respiratory symptoms requiring hospitalisation and/or a course of oral corticosteroids and/or antibiotics (either prescribed or self administered). Patients are advised to contact the investigator if they experience an increase in respiratory symptoms for 2 consecutive days, and where possible, prior to commencing on a course of corticosteroids or antibiotics.
- An ALT / AST level ≥ 3xULN confirmed by a repeat test, or any pattern of liver function test (LFT) abnormalities giving Investigator or AZ cause for concern
- Evidence of haemolysis as evidenced by the following:
  - Reticulocyte count of over 5%, **and**

  At least two of the following:
  - a drop in haemoglobin below 10.5 g/dl
  - decrease of 25% in haptoglobin. (The value post-dose to be compared with the average of two pre-dose values. The value should also be 15% below LLN).
  - increase of 25% in unconjugated bilirubin or lactate dehydrogenase (LDH). (The value post-dose to be compared with the average of two pre-dose values. The value should also be 15% above ULN).

- A platelet count of below 80 x 10^9/L confirmed by a repeat test (in the absence of obvious platelet clumping in a peripheral blood film).

Specific reasons for discontinuing a patient from the genetic research are:

- Withdrawal of consent for genetics research. A patient may withdraw from this genetic research at any time, independent of any decision concerning participation in other aspects of the main study described in this protocol. Voluntary discontinuation by the patient will not prejudice further treatment.

Stopping criteria for the whole study

- Two or more patients on active treatment are discontinued because of either an ALT/AST level ≥3 x ULN **or** any pattern of LFT abnormalities giving cause for concern

- Two or more patients on active treatment are discontinued because of evidence of haemolysis.

- Two or more patients on active treatment are discontinued because of a platelet count of below 80 x 10^9/L.

3.3.5.2 Procedures for discontinuation

Patients who discontinue should always be asked about the reason(s) for their discontinuation and the presence of any adverse events. If possible, they should be seen and undergo the procedures and assessment of the Follow-up visit (Visit 5). Adverse events should be followed up; diary cards and investigational products should be returned by the patient.

3.3.5.3 Procedures for handling incorrect enrolled patients

Patients not meeting the inclusion/exclusion criteria for a study should under no circumstances be enrolled. There can be no exceptions to this rule.

Where patients not meeting the study criteria are randomised in error or where patients subsequent to randomisation fail to meet the criteria for the study, the Principal Investigator should be informed, and the patient withdrawn from the study as soon as possible, unless there are sufficient ethical or safety reasons that the patient should remain in the study. The decision on how to proceed will be documented.

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3.3.5.4 Procedures for discontinuation from genetic aspects of the study

Patients who discontinue from the study should always be asked specifically whether they are withdrawing or continuing their consent for this genetic research. It must be established whether the patient:

- Agrees to the genetic sample and any DNA extracted from the sample being kept for genetic research in the future.

- Withdraws consent for the sample to be kept for genetic research in the future and wishes the sample to be destroyed. Destruction of the sample (or the DNA extracted from the sample) will only be possible so long as the particular sample is traceable. In the event that genetic research has already been performed, AstraZeneca will retain the results and associated data for regulatory reasons but these will not be used in any subsequent analyses.

The principal investigator is responsible for providing written notification to AstraZeneca of any patient who has withdrawn consent for the use of the sample taken for genetic research. AstraZeneca will provide written confirmation to the investigator of the actions taken with the sample, which must be filed in the investigator study file.

3.4 Treatments

3.4.1 Identity of investigational product and comparators

AZD9668 will be provided as a tablet for oral administration and with a matching placebo. Tablets of AZD9668 will be provided in the strength of 30 mg.

Table 2 Identity of investigational product

Investigational product	Dosage form and strength	Manufacturer	Formulation number	Batch number
AZD9668	Tablet, 30 mg	AstraZeneca R&D, [redacted]	1756-1	Will be detailed in the Clinical study report (CSR)
Placebo AZD9668	Tablet, not applicable	AstraZeneca R&D, [redacted]	P1756-0	Will be detailed in CSR

The composition of the tablets is 30 mg AZD9668 (corresponding to 39.5 mg AZD9668 tosylate).

Excipients: cellulose microcrystalline, calcium hydrogen phosphate dihydrate, crospovidone, sodium laurilsulfate, sodium stearyl fumarate.

The drug product consists of white to off white, plain oblong, biconvex tablet. The nominal tablet weight is 400 mg.

Placebo tablets to match AZD9668 tablets contain the following excipients: cellulose microcrystalline and sodium stearyl fumarate

3.4.2 Doses and treatment regimens

In the current study the daily dose will be 120 mg (i.e. 60 mg b.i.d), except for days 1 and 14 when patients will receive only the morning dose to allow assessment of PK. This dose has been chosen as a dose that is predicted to achieve trough steady state therapeutic plasma concentrations of 19 nM (predicted from an in vivo mouse acute lung injury model) in all patients, whilst ensuring that the predicted geometric mean $C_{max,ss}$ and $AUC_{(0-24)ss}$ are below the HELC limits. The estimated margins to the HELC limits are 2.6 and 1.4 for $C_{max,ss}$ and $AUC_{(0-24)ss}$, respectively, in the fasted state. If a dose of 60 mg b.i.d is not tolerated by an individual patient then his/her dose may be halved to 30 mg b.i.d for the remainder of the study, at the investigator's discretion. The population PK model predicts that this will still achieve trough steady state plasma concentrations above 19 nM in all patients.

For morning doses administered on Days 1, 7 and 14, patients will be fasted from midnight the night before dosing except water for thirst. The patients may consume water up to 1 hour before dosing. The dose will be administered with 240 mL of water, and the patients must drink all the water. The patients must not consume any further fluids or food for a minimum of 2 hours post dose. At this time the patients will be allowed a light breakfast. Fasting the patients prior to receiving their morning dose on days when they are having ECG measurements ensures that the ECG measurements can be taken when C_{max} is maximised. There is no requirement to restrict food consumption before or immediately after administration of the remaining doses. These doses can be administered with approximately 100 mL water.

The morning and evening doses will be filled individually in high-density polyethylene (HDPE) bottles with a child resistant cap according to the random scheme. All bottles for morning and evening dose for 14 days will be packed in a white box for each patient.

At Visit 2, the required number of bottles needed for the morning and evening dose until Visit 3 will be handed over to the patient after the 4^{th} dose has been taken in the CPU.

At Visit 3, the required number of bottles until Visit 4 will be handed over to the patient after the morning dose has been administered.

At Visit 4, the patient will be administered the morning dose in the CPU by the staff.

3.4.3 Labelling

The packaging and labelling will be performed by [redacted] AstraZeneca R&D [redacted]. All supplies and labels will be prepared in accordance with Good Manufacturing Practice (GMP) and local regulatory guidelines. The labels will fulfil GMP Annex 13 requirements for labelling.

3.4.4 Storage

All investigational products must be kept in a secure place under appropriate storage conditions. A description of the appropriate storage & shipment conditions are specified on

the investigational product label. The storage location will be locked and only accessible to authorised site personnel.

3.4.5 Accountability

The medication provided for this study is for use only as directed in the protocol. All unused study drugs will be accounted for and destroyed/returned appropriately by CPU personnel. The study site personnel will account for all drugs dispensed and returned. Certificates of delivery and return must be signed.

The investigator is responsible for maintaining study drug accountability at site.

3.5 Method of assigning patients to treatment groups

Informed consent will be obtained before enrolment and the patients identified with an enrolment number.

Patient eligibility will be established before treatment randomisation. Patients will be randomised strictly sequentially, as patients are eligible for randomisation. Patients fulfilling the eligibility criteria will be assigned randomisation codes (unique subject numbers), starting with 001. Numbers will be allocated in ascending order. If a patient discontinues from participation in the study, the patient number will not be re-used, and the patient will not be allowed to re-enter the study.

3.6 Blinding and procedures for unblinding the study

3.6.1 Methods for ensuring blinding

AZD9668 and placebo tablets are manufactured to appear similar in size, shape, weight and colour.

3.6.2 Methods for unblinding the study

Individual treatment codes, indicating the treatment randomisation for each randomised patient, will be available to the investigator(s) or pharmacists at the study centre.

A copy of the randomisation scheme will be made available to the pharmacist, the analyst of AZD9668 in sputum and the PK analyst to enable analysis of samples from patients who have received active treatment to be prioritised. This documentation will be placed in a secure location until the end of the study.

The treatment code must not be broken except in medical emergencies when the appropriate management of the patient necessitates knowledge of the treatment randomisation. The codes may also be unblinded, after consultation with the Drug Safety Physician at AstraZeneca, if 2 or more patients meet the discontinuation criteria as specified in Section 3.3.5.1. The investigator(s) must document and report to AstraZeneca any breaking of the treatment code. AstraZeneca retains the right to break the code for SAEs that are unexpected and are suspected to be causally related to an investigational product and that potentially require expedited reporting to regulatory authorities.

Treatment codes will not be broken for the planned analyses of data until all decisions on the evaluability of the data from each individual patient have been made and documented.

3.7 Pre-study, concomitant and post-study treatments

Patients are allowed to continue long-Acting muscarinic antagonist (LAMA), long-acting beta agonists (LABA), LABA/ICS (inhaled corticosteroids) combinations unchanged throughout the study if they are already on these. They would only need to withhold LAMA/LABA for 12 hours, and short-acting beta agonists (SABA) for 4 hours before the days on which they have their pulmonary function tests. After they have had the test they could take their normal dose of the medication. Other respiratory medications could be continued unchanged.

The following drugs (substrates for CYP2C9) will not be allowed throughout the dosing period:

- Anti-diabetics/hypoglycaemics
- Anti epileptics/anticonvulsants
- Warfarin
- Fluvastatin
- Celecoxib
- High dose continuous non-steroidal anti-inflammatory drugs (NSAIDs)
- Torsemide
- Amitryptiline
- Fluoxetine

Any other drugs including inhaled and oral respiratory drugs could be allowed provided their doses have been stable for at least 4 weeks before enrolment and the doses remain unchanged throughout the study. SABA and/or SAMA may be taken as needed for symptomatic control of breathlessness.

Other medication, which is considered necessary for the patient's safety and well-being, may be given at the discretion of the investigator. The administration of all medication (including investigational products) must be recorded in the appropriate sections of the [redacted]™.

3.8 Treatment compliance

Treatment compliance will be checked by counting the returned medication on Day 7 and 14. In addition for the patients receiving active drug, the blood concentration of AZD9668 will serve as control.

Patients will be asked to return their unused medication and all packaging (even if empty) to the study centre at each visit for reconciliation purposes.

4. MEASUREMENTS OF STUDY VARIABLES AND DEFINITIONS OF OUTCOME VARIABLES

4.1 Primary variable

For description of primary variables refer to Section 4.5, pharmacokinetic, and Section 4.7, safety variables and measurement.

4.2 Screening and demographic measurements

- Demographic data, i.e. date of birth, sex, height, weight, BMI, race
- A standard medical and surgical history and a physical examination including the cardiovascular and respiratory systems
- Use of nicotine
- Concomitant medication
- Standard clinical chemistry (including FSH at Visit 1 for female patients) and haematology assessments and a mid-stream urine sample for urinalysis, and drugs of abuse screen (and pregnancy test in female patients)
- A resting blood pressure and pulse measurement (supine).
- An alcohol screen (urine test)
- A 12-lead ECG
- Hepatitis B surface antigen, Hepatitis C antibody and HIV status
- Spirometry (lung function)

4.3 Patient-Reported Outcomes (PROs)

Not applicable.

4.4 Health Economic measurements and variables

Not applicable.

4.5 Pharmacokinetic measurements and variables

For timing of individual samples refer to the detailed study plan (Table 1).

The methods for collection of biological samples and derivation of pharmacokinetic variables are presented below in Sections 4.5.1 and 4.5.2

4.5.1 Collection of biological samples

Blood samples (3 ml K_2EDTA**)** for determination of AZD9668 in plasma will be taken at the times presented in the detailed study plan (Table 1).

Blood samples will be collected, labelled and shipped as detailed in the laboratory handbook. The date and time of collection will be recorded.

Urine will be passed into a specially provided container at the times presented in Table 1. Urine samples will be collected, handled, labelled and shipped as detailed in laboratory handbook. The date and time of collection will be recorded.

Samples should be stored at –18°C or below, and analysed within the timeframe after collection for which the stability in the samples has been validated and found acceptable. Results from analyses stored longer than the period stated will not be reported.

Aliquots of sputum for AZD9668 analysis will be collected, handled, labelled and shipped as detailed in the laboratory handbook. The date and time of collection will be recorded.

Samples should be shipped to the appointed bioanalytical laboratory at pre-determined dates during the course of the study.

The methods used will be referred to in the clinical study report.

Samples will be disposed of within one month after the clinical study report has been finalised.

4.5.2 Drug concentration measurements, and derivation or calculation of pharmacokinetic parameters

Samples for determination of AZD9668 in plasma and urine will be analysed by [redacted], on behalf of [redacted], AstraZeneca R&D [redacted], using validated methods of liquid chromatography and mass-spectrometry (LC-MS/MS). The lower limit of quantification (LLOQ) of AZD9668 in plasma and urine will be 1.00 nM and 0.2 µM, respectively.

The PK parameters will be calculated using non-compartmental analysis. The pharmacokinetic parameters will include:

Day 1: $AUC_{(0\text{-}t)}$, $AUC_{(0\text{-}24)}$, AUC, $t_{1/2}$, CL/F, V_z/F, $C_{24,}$ CL_R, A_e and F_e(%)

Day 7: $C_{min,ss}$

Day 14: $C_{min,ss}$, $C_{max,ss}$, $t_{max,ss}$, $AUC_{(0-t),ss}$, $AUC_{(0-24),ss}$, $C_{24,ss}$, AUC_{ss}, $t_{1/2ss}$, CL/F_{ss}, V_z/F_s, R_{ac}, CL_R, $A_{e,ss}$ and $F_{e,ss}$(%)

The accumulation ratio (R_{ac}) will be calculated using the following equation:

$AUC_{(0-24),ss}$ Day 14/$AUC_{(0-24)}$ Day 1.

An investigation of the AZD9668 concentrations in sputum will be conducted using a method of liquid chromatography and mass spectrometry (LC-MS/MS) based on that validated for human plasma.

The AZD9668 sputum analysis method will not be formally validated, but will be supported by investigations into the recovery and stability of AZD9668 in this matrix and each batch of samples will include QC samples to demonstrate the performance of the method.

Measurement of AZD9668 concentrations in sputum will be analysed by [redacted], AstraZeneca R&D [redacted].

4.6 Efficacy and pharmacodynamic measurement and variables

4.6.1 Efficacy parameters

Not applicable.

4.6.2 Pharmacodynamic parameters

4.6.2.1 Induced sputum weight, neutrophils, hydroxyproline

Handling and processing of induced sputum will be performed according to validated methods. The handling, storing, shipping, method of analysis and prioritised order of analysis for the different parameters will be detailed in the laboratory handbook.

4.6.2.2 Biomarkers in serum, plasma and urine

Blood and urine samples for determination of serum amyloid A, hsCRP, cytokines (including but not limited to TNF-α and IL-8), desmosine in plasma and desmosine in urine will be taken at the times given in the study plan (Table 1). The handling, storing, shipping, method of analysis and prioritised order of analysis for these different parameters will be detailed in the laboratory handbook.

~~4.6.2.2~~4.6.2.3 BronkoTest© diary card

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The BronkoTest© sputum colour test (BronkoTest©, Hampton Hill, UK) is used to characterise a COPD patient's normal chest-related symptoms and the changes that occur during an exacerbation. It documents the 3 key symptoms of such episodes, namely increased breathlessness, new sputum production or increased volume, and the purulent nature of the sputum.

In the dairy card, the following items will be filled in daily by the patients: Peak flow measurement, night-time symptoms, description of breathing, type, colour and amount of

sputum and as well associated symptoms about well-being, frequency of coughing, occurrences of chest pain and cold or flu. In addition, the treatment and management of the COPD symptoms and intake of any antibiotics will be documented.

For Peak flow measurement, the subject will be provided with and instructed to use the APP® Mini-Wright Peak Flow Meter "Standard" (Beckmann, Seefeld, Germany).

During the Screening visit, the patients will receive the BronkoTest© diary card and the Peak flow meter and be instructed how to use the Peak flow meter and how to fill in the BronkoTest© diary card.

4.7 Safety measurements and variables

The methods for collecting safety data are described below.

4.7.1 Adverse events

4.7.1.1 Definitions

The definitions of adverse events (AEs), serious adverse events (SAEs) and other significant adverse events (OAEs) are given below. It is of the utmost importance that all staff involved in the study are familiar with the content of this section. The principal investigator is responsible for ensuring this.

Adverse event

An adverse event is the development of an undesirable medical condition or the deterioration of a pre-existing medical condition following or during exposure to a pharmaceutical product, whether or not considered causally related to the product. An undesirable medical condition can be symptoms (eg, nausea, chest pain), signs (eg, tachycardia, enlarged liver) or the abnormal results of an investigation (eg, laboratory findings, electrocardiogram). In clinical studies, an AE can include an undesirable medical condition occurring at any time, including run-in or washout periods, even if no study treatment has been administered.

Serious adverse event

A serious adverse event is an AE occurring during any study phase (ie, run-in, treatment, washout, follow-up), and at any dose of the investigational product, comparator or placebo, that fulfils one or more of the following criteria:

- Results in death
- Is immediately life-threatening
- Requires in-patient hospitalisation or prolongation of existing hospitalisation
- Results in persistent or significant disability or incapacity
- Is a congenital abnormality or birth defect

- Is an important medical event that may jeopardise the patient or may require medical intervention to prevent one of the outcomes listed above.

The causality of SAEs (ie, their relationship to study treatment) will be assessed by the investigator(s), who in completing the relevant case report form must answer "yes" or "no" to the question "Do you consider that there is a reasonable possibility that the event may have been caused by any of the following – study medication – other medication?" For further guidance on the definition of a SAE and a guide to the interpretation of the causality question, see Appendix B to the Clinical Study Protocol.

Note that SAEs that could be associated with any study procedure should also be reported. For such events the causal relationship is implied as "yes".

Other Significant Adverse Events (OAE)

OAEs will be identified by the Study Delivery Team Physician in consultation with the appropriate Global Drug Safety Physician during the evaluation of safety data for the Clinical Study Report. Significant adverse events of particular clinical importance, other than SAEs and those AEs leading to discontinuation of the patient from study treatment, will be classified as OAEs. Examples of these are marked haematological and other laboratory abnormalities, and certain events that lead to intervention (other than those already classified as serious), dose reduction or significant additional treatment. For each OAE, a narrative may be written and included in the Clinical Study Report.

4.7.1.2 Recording of adverse events

AEs will be recorded in the CRF from consent (Visit 1) until Visit 5. Any AEs remaining unresolved at Visit 5 should be recorded as "ongoing" in the CRF. Where appropriate, these should be followed as long as medically indicated but without further recording in the CRF. AstraZeneca retains the right to request additional information judged necessary, for any patient with ongoing AE(s)/SAE(s) at the end of the study.

The following variables will be recorded for each AE: AE description, start and stop date, intensity, action taken, outcome, investigator's causality rating (yes or no), and whether the AE meets criteria for a SAE or not.

The following levels of intensity will be recorded

- mild (awareness of sign or symptom, but easily tolerated)
- moderate (discomfort sufficient to cause interference with normal activities)
- severe (incapacitating, with inability to perform normal activities)

It is important to distinguish between serious and severe AEs. Severity is a measure of intensity whereas seriousness is defined by the criteria in Section 4.7.1.1. An AE of severe intensity need not necessarily be considered serious. For example, nausea that persists for

several hours may be considered severe nausea, but not a SAE. On the other hand, a stroke that results in only a limited degree of disability may be considered a mild stroke but would be a SAE.

AEs will be collected on an ongoing basis All AEs spontaneously reported by the subject or reported in response to open questions from the study personnel: "Have you had any health problems since the previous visit?" or "Have you had any health problems since you were last asked" or revealed by observation will be collected and recorded in the CRF. The first AE questioning will occur at Visit 2.

Signs and symptoms present at Visit 1 will be recorded as 'ongoing pre-dose'. When the severity or seriousness of any pre-dose signs or symptoms changes this will be recorded as a new AE.

AEs and any abnormalities in laboratory tests, vital signs or physical examination of clinical relevance will be followed up until resolution or no further improvement is expected in the opinion of the Investigator. Patients who drop out during the study for reasons other than discontinuation of study drug will be followed up for the planned duration of the study. Abnormalities in laboratory tests and vital signs will be recorded and reported on the laboratory reports and CRFs respectively. Abnormalities in these variables will be identified, analysed and reported in the Clinical Study Report. Laboratory results that constitute a SAE or lead to discontinuation of the study drug will be recorded and reported as an AE. In addition, any abnormality in laboratory tests which meets the stopping criteria as described in Section 3.3.5, will be recorded as an AE.

Should an overdose (accidental or deliberate) occur, it must be reported in accordance with the procedures described in Section 9.3, Procedures in case of overdose, regardless of whether the overdose was associated with any symptom or not. All symptoms associated with the overdose should be reported as AEs.

Should a pregnancy occur, it must be reported in accordance with the procedures described in Section 9.4 - Procedures in case of pregnancy. Pregnancy in itself is not regarded as an AE unless there is a suspicion that an investigational product may have interfered with the effectiveness of a contraceptive medication.

If patients annotate their diary card or questionnaires with comments, these will be reviewed by the study centre staff in consultation with the patient, in case the annotation represents an AE.

4.7.1.3 Reporting of serious adverse events

Investigators and other site personnel must inform appropriate AstraZeneca representatives of any SAE that occurs in the course of the study within 1 day (i.e. immediately but no later than the end of the next business day) of when he or she becomes aware of it.

Follow-up information on SAEs must also be reported by the investigator within the same time frames.

If a non-serious AE becomes serious, this and other relevant follow-up information must also be provided to AstraZeneca within 1 day as described above. For a non-serious AE that become serious but which is not fatal or life-threatening a report should be received within 5 days.

The AstraZeneca representative will work with the investigator to compile all the necessary information and ensure that the appropriate AstraZeneca Drug Safety Department receives a report by day 1 for all fatal and life-threatening cases and by day 5 for all other SAEs.

All SAEs have to be reported, whether or not considered causally related to the investigational product or to the study procedure(s). All SAEs will be recorded in the pCRF. The investigator and/or Sponsor are responsible for informing the Ethics Committee and/or the Regulatory Authority of the SAE as per local requirements. For studies in countries implementing the EU Clinical Trials Directive, this will be taken care of by AstraZeneca (see Section 8.1).

4.7.2 Laboratory safety measurements and variables

4.7.2.1 Methods of assessment

Blood and urine samples for determination of clinical chemistry, haematology and urinalysis parameters, will be taken at the times given in the study plan (Table 1). The date of collection will be reported in [redacted]™.

Blood and urine collection for all safety parameters will be performed according to validated methods of AstraZeneca and [redacted] Information for handling, labelling, storing and shipping will be provided in the laboratory handbook for safety blood and urine analyses.

The following laboratory variables ~~for safety laboratory~~ will be measured:

Clinical chemistry	**Haematology**
P-Creatinine	B-Haemoglobin
~~P-Total and unconjugated bilirubin~~ P- Total bilirubin [a]	~~B-Leucocyte count~~B-Leucocyte count (absolute and differential)
P-Alkaline phosphatase	B-Red cell count (RBC)
P-Aspartate aminotransferase (AST)	B-Haematocrit
P-Alanine aminotransferase (ALT)	B-Mean cell volume (MCV)
P-Albumin	B-Mean cell haemoglobin (MCH)
P-Potassium	B-Mean cell haemoglobin concentration (MCHC)
P-Calcium, total	B-Platelet count
P-Sodium	B-Reticulocyte count
P-Cholesterol	
P-Triglycerides	**Urinalysis**

P-Urea	U- Haemoglobin (RBC)
P-Gamma Glutamyltransferase (GGT)	U-Protein
P-Creatinine phosphokinase (CPK) [b]	U-Glucose
P-LDH	Pregnancy / FSH
P-Glucose (fasting)	βhCG / FSH as appropriate
P-Free thyroxine (T4) [c]	
P-Thyroid stimulating hormone (TSH) [c]	
P-Haptoglobin	
Virology [d]	

a If elevated unconjugated bilirubin will be measured
b If elevated, P-CPK-MB will be analysed (will not be databased)
c Included at Visit 1 only
d Hepatitis B and C and HIV status will be checked at Visit 1 only

The respective laboratory reference ranges will be provided by the local laboratory and filed in the Investigator Site File (ISF).

These laboratory tests will be performed at the [redacted] using validated standard methods according to the applicable laboratory SOP. Laboratory data will be transmitted electronically from the [redacted] to the study site.

4.7.2.2 Sputum for quantitative bacteriology

Spontaneous sputum samples for quantitative bacteriology will be taken at the times given in the study plan (Table 1). These will be performed according to the guidelines established at the local laboratory. Information for handling, labelling, storing and shipping of these sputum samples will be provided in the laboratory handbook.

~~4.7.2.2~~4.7.2.3 Derivation or calculation of outcome variables

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All individual values and changes to baseline of the laboratory parameters will be listed and summarized descriptively. Result of Virology and Drug screen including alcohol will only be listed.

Abnormal findings for laboratory variables should only be reported as AEs if they are accompanied by clinical symptoms, constitute SAEs or lead to discontinuation of treatment with the investigational product.

4.7.3 Vital signs, ECG and physical examination

All assessments will be performed at time points indicated in the flow chart (Table 1).

4.7.3.1 Methods of assessment

Vital signs

Vital signs comprise the assessment of systolic and diastolic blood pressure, pulse, respiratory rate and body temperature. Assessments of vital signs resulting in clinically significant abnormal values will be repeated, in order to exclude an erroneous assessment. Controls will also be necessary if implausible measurements occur, e.g., significant deviations from previous assessments. Individual results will be classified as 'normal', 'abnormal with no clinical significance' and 'abnormal with clinical significance'. Abnormal values 'with no clinical significance' may be considered clinically significant if they are accompanied by clinical symptoms such as dizziness, tiredness, nausea or vomiting etc.

All measurements of vital signs will be done according to the applicable [redacted] SOP.

Twelve-lead ECG

Twelve-lead ECGs will be recorded in accordance with local procedure.

A resting ECG will be recorded at Visit 1 for confirmation of study eligibility and at Visit 5 for follow-up safety assessment. Further ECG recordings will be performed according to the study plan (Table 1). ECGs will be recorded in the supine position after the patient has rested in this position for at least 10 minutes. Only overall evaluation will be captured in [redacted]™. Any abnormalities (including QTc values) should be reviewed by a cardiologist or an appropriately qualified person.

The original ECGs and variables must be signed and dated and stored in the patient's medical record as source data.

Telemetry

Ambulatory cardiac monitoring will be performed using CardioMem CM3000 recorders (Getemed, Germany) from 24 hours pre-dose to 24 hours post first dose at Visit 2. The Investigator or delegate will review and evaluate the recordings before dosing, and before discharge. A print-out will be signed and dated by the Investigator and filed with the CRF.

Physical examination

A full physical examination will be performed to ensure suitability according to the inclusion and exclusion criteria at the Screening visit. The physical examination comprises a routine medical examination including gross neurological assessments. Measurement of body weight and height will be done as well.

The physical examination will be repeated as the follow-up visit and any new findings should be recorded as adverse events.

4.7.3.2 Derivation or calculation of outcome variables

All individual values and changes to baseline will be listed and summarized descriptively.

Abnormal findings for vital signs and ECGs should only be reported as AEs if they are accompanied by clinical symptoms, constitute SAEs or lead to discontinuation of treatment with the investigational product.

4.7.4 Other safety measurements and variables

Lung function tests will be recorded whilst the subject is in a sitting position at the time points specified in the study flow chart (Table 1).

The lung function tests will comprise:

- Forced expiratory volume in one second (FEV_1)
- Forced vital capacity (FVC)
- Slow vital capacity (SVC)
- Inspiratory capacity (IC)

Spirometry will be performed with the same mobile bi-directional digital computerised spirometer (Masterscope®, Jaeger, Höchberg, Germany) with software Labman, version 4.53, for each subject throughout the study according to the European Respiratory Standards (ERS).

All lung function tests will be repeated, until three technically acceptable measurements have been made and the maximum of these three measures will be recorded in the CRF. Post-bronchodilator values will be obtained 20 minutes after inhalation of 400µg of salbutamol. The details of the procedure for the performance of the laboratory tests will be given in the laboratory handbook.

4.8 Volume of blood sampling and handling of biological samples

The total volume of blood that will be drawn from each patient in this study is as follows:

Table 3 Volume of blood to be drawn from each patient

Assessment		Sample volume (mL)	No. of samples	Total volume (mL)
TNFα desmosine and IL-8 (plasma)		10.0 mL	3	30.0
Amyloid-A and hs-CRP (serum)		2.7 mL	4	10.8
PK		3.0 mL	25	75.0
Virology		2.6 mL	1	2.6
Pregnancy/FSH		2.7 mL	1	2.7
Safety	Clinical chemistry	7.5 mL	6	45.0
	Haematology	2.7 mL	6	16.2
Genotyping		9.0 mL	1	9.0

Table 3 Volume of blood to be drawn from each patient

Assessment	Sample volume (mL)	No. of samples	Total volume (mL)
Total			191.3

4.8.1 Analysis of biological samples

4.8.1.1 Clinical chemistry samples

The analyte stability limits defined by [redacted] will be applied to all analyses performed on behalf of AstraZeneca. [redacted] will not analyse samples that fall outside these stability limits. Analytical data will not be reported if found to have been derived from a sample that fell outside these stability limits. The standards of procedure followed by [redacted] may be amended in accordance with its Standard Operating Procedures. [redacted] will inform AstraZeneca of the stability limits relevant to this study before the first patient gives informed consent to take part in the study.

If [redacted] chooses to sub-contract the analytical work to another laboratory, [redacted] must assure itself and provide assurance to AstraZeneca that the other laboratory will apply defined stability limits to all analyses performed on behalf of AstraZeneca. Samples falling outside these limits must not be analysed or data reported. The other laboratory will inform AstraZeneca of the stability limits relevant to this study before the first patient gives informed consent to take part in the study.

4.8.1.2 Pharmacokinetic samples

The long-term stability of the analyte in plasma and urine has been documented in method validation produced by [redacted]. Results from analyses of samples stored longer than the time period for which stability has been demonstrated should not be reported unless complementary analyte stability data is acquired and amended to the relevant method validation report. Documentation of the time period for which stability has been demonstrated is available at [redacted].

4.9 Genetic measurements and co-variables

4.9.1 Collection of samples for genetic research

Patients will provide a blood sample as per the inclusion criteria and visit schedule.

A single venous blood sample 9 mL will be collected into a polypropylene tube containing ethylenediamine tetra-acetic acid (EDTA) and gently inverted a minimum of 5 times to mix thoroughly. Tubes will be labelled with the protocol study number, centre number, enrolment code and/or randomisation number and date of sample collection. No personal identifiers (patient name, initials or date of birth) will be placed on the tube or accompanying documentation. A record of the date of the patient consent to the genetic research and the date of the blood sample collection will be recorded in the appropriate section of [redacted]™.

Genotype is a stable parameter; therefore if for any reason the blood sample is not drawn at Visit 2, it may be taken at any visit until the last study visit. The genetic blood sample should ideally be drawn through the same cannula used to draw blood samples required for the main study.

4.9.1.1 Sample processing and shipping

Samples will be frozen (-20°C or below) and transported to the relevant DNA extraction laboratory within one month of collection and must remain frozen at all times.

Where possible samples should be shipped in batches and shipment should be coordinated with the receiving site to ensure that samples arrive within working hours. A requisition sheet, detailing the protocol study number, centre number, enrolment code and/or randomisation code and date of sample collection, should accompany the shipment.

4.9.1.2 Storage and coding of DNA samples

The processes adopted for the coding and storage of samples for genetic analysis are important to maintain patient confidentiality.

For all samples irrespective of the type of coding used the DNA will be extracted from the blood sample. The DNA sample will be assigned a unique number replacing the information on the sample tube. Thereafter, the DNA sample will be identifiable by the unique DNA number only. The DNA number will used to identify the sample and corresponding data at the AstraZeneca genetics laboratories, or at the designated contract laboratory. No personal details identifying the individual will be available to any AstraZeneca employee working with the DNA.

'The blood samples and data for genetic analysis in this study will be coded. Each blood sample will be labelled with the study number and subject number. Only the investigator will be able to link the blood sample to the individual subject. The sample and data will not be labelled with a personal identifier. The link between the subject enrolment/randomisation code and the DNA number will be maintained

All DNA samples will be stored under secure conditions with restricted access at AstraZeneca. The blood, DNA samples or data derived from the samples may be made available to groups or organisations working with AstraZeneca on this study or as part of the development drug project. However, the samples and any results will remain the property of AstraZeneca at all times. AstraZeneca will not give blood, DNA samples or data derived from the samples to any other parties, except as required by law.

4.9.1.3 Summary of genetic assessments and analysis

The purpose of the genetic research is to generate data for use in future retrospective analyses. Future analyses will explore genetic factors that may influence the disposition, efficacy, safety and tolerability to AZD9668 and/or susceptibility to or prognosis of COPD under investigation in this protocol. The results of the genetic research will not form part of the

clinical study report for this study. The results may be pooled with genetic data from other studies on AZD9668 to generate hypotheses to be tested in future studies.

4.9.1.4 Derivation or calculation of genetic parameters

The number of patients who will agree to participate in the genetic research is unknown. It is therefore not possible to establish whether sufficient data will be collected to allow a formal statistical evaluation or whether only descriptive statistics will be generated. A statistical analysis plan will be prepared where appropriate.

5. DATA MANAGEMENT

~~5.1.1~~5.1 Data collection methods and data validation at study site

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~~5.1.1.1~~5.1.1 Data flow

[redacted]™ system is an electronic source data capturing and information management system. The system combines all aspects of source data capturing with process control and study management. All clinical and laboratory data, except those which are paper based, will be collected by the eSource system [redacted]™. Only paper-based data will be subject to data entry. For electronic data, no data entry will be performed. Data entered on paper CRF should be recorded legibly in blue or black ballpoint pen. Correction fluid or covering labels must not be used.

The responsible monitor will check data at the monitoring visits to the study site. The Investigator will ensure that the data collected are accurate, complete and legible. Data will be validated within [redacted]™ by the investigator and the study monitor before being exported. Any changes made during validation will be documented with a full audit trail within [redacted]™.

[redacted] designs and builds a database in iVal to enter the paper-based data for adverse event and concomitant medication. The database and entry screens will be built and validated following a formal, documented verification plan and the system will be tested against the specification for the entry system. The setup will be controlled by test data entry. A standardised setup of SAS table sets in AstraZeneca specification will be done by the SAS programmer. Verification of the setup will be performed with dummy and test data.

The [redacted] data manager will develop a DVS according to the protocol, the data dictionary of the iVal database for paper based-data and the description of the SAS table set for electronic data. Computerised checks to be performed on the data imported or entered into the databases will be defined in this document. The DVS will be sent to AstraZeneca or representative for review and approval.

Paper-based data will be sent to [redacted] data management and entered into iVal. These data will be exported via the module iSAS later on and merged with the SAS tables in AstraZeneca format. The SAS table sets will be used for verification of the electronic study

data following the specifications listed in the DVS. Only trained staff will have access to the databases. Every change will be fully audit-trailed.

The [redacted]™ data files will be imported into SAS table sets. The imported data checked by the related verification programmes specified in the DVS. Data will be changed only in a controlled, audit-trailed, environment. After these data are considered as clean, they will be mapped into AstraZeneca data structure.

All study related electronic source data captured in [redacted]™ will be transferred from [redacted]™ via [redacted] to [redacted] Data Management after monitoring. These data will be imported into the SAS table set. Paper based data will be sent to Data Management after they have been monitored. [redacted] Data Management reviews, logs and files them.

Any missing, impossible or inconsistent recordings will be referred back to the Investigator using a data query form and be documented for each individual subject before clean file status is declared.

In the case of genotypic data, only the date the subject gave consent to participation in the genetic research and the date the blood sample was taken from the subject will be recorded in [redacted]™.

The genotypic data generated from the study will be stored in the AstraZeneca LIMS database or other appropriate system. This database is a secure database, which is separate the database used for the main study. Some or all of the dataset from the main study may be duplicated within the AstraZeneca LIMS database for exploratory genetic analysis

5.1.2 Study Data Management Plan (DMP)

The Study Data Management Plan (DMP) will describe the methods used to collect, check and process clinical data in detail. It will also clarify the roles and responsibilities for the different functions and personnel involved in the data management process. The DMP will also describe the data flow and timelines within the study.

5.1.3 Coding tools and dictionaries

Adverse events (AEs) and diagnoses from medical history will be classified according to the terminology of Medical Dictionary for Regulatory Activities (MedDRA). Concomitant medications will be classified according to the AZDD (AstraZeneca Drug Dictionary), the Anatomical Therapeutic Chemical (ATC) system and the Committee of Proprietary Medicinal Products (CPMP) route of administration dictionary. All coding will be performed by [redacted]

5.2 Reporting of genotypic results

Results from any genetic research performed will be reported separately from the clinical study report. AstraZeneca will not provide individual genotype results to patients, their family members, any insurance company, an employer, clinical study investigator, general physician

or any other third party, unless required to do so by law. The patient's DNA will not be used for any purpose other than those described in the study protocol.

Individual patients will not be identified in any report or publication resulting from this work. The data and results of this study may be reviewed with collaborators and published, but neither the patient's name nor any other personal identifiers will appear in any publication or report.

6. STATISTICAL METHODS AND DETERMINATION OF SAMPLE SIZE

6.1 Statistical evaluation – general aspects

A comprehensive Statistical Analysis Plan (SAP) will be prepared before database lock and unblinding of the data in order to avoid any potential bias due to knowledge of the treatment received. It will detail analyses to be performed and summaries to be produced for the Clinical Study Report (CSR).

All statistical summaries and analyses will be performed by [redacted] using SAS® (Statistical Analysis system, SAS-Institute, Cary NC, USA).

Pharmacokinetic parameters will be derived by non-compartmental analysis using WinNonLin Pro Version 5.0 or higher version.

6.2 Description of outcome variables in relation to objectives and hypotheses

This is an exploratory study and no hypotheses will be tested.

6.3 Description of analysis sets

The safety analysis set comprises all patients randomised into the study and receiving at least one dose of study medication. Patients will be analysed according to the treatment they actually received.

The PK analysis set is a subset of the safety analysis set and will only include patients who showed evaluable data from Day 1 and Day 14.

6.4 Method of statistical analysis

6.4.1 General analysis methods

Data will be summarised by treatment using descriptive statistics. All data (including derived data) will be listed, and will be summarised and plotted as appropriate, further details of the output to be produced will be contained in the SAP.

In addition biomarker data will be analysed using Analysis of variance The exploratory data may be explored further, and this information may be included in the CSR. Demographic and baseline variables (medical and surgical history details and abnormal results of physical examinations) will be summarised and listed.

The safety analysis set will be used as the primary analysis data set for the reporting of safety data; while the analysis of PK data will be based on the pharmacokinetic analysis set.

No imputation method for missing values will be used; data will be evaluated as observed.

6.4.2 Pharmacokinetic concentrations and variables

The pharmacokinetic analyses will be performed by or under the guidance of [redacted] International GmbH.

For descriptive statistics of plasma concentrations

- if, at a given time point, 50% or less of the plasma concentrations are non-quantifiable (NQ), the geometric mean (gmean), CV, geometric mean ± SD, arithmetic mean and SD will be calculated by substituting the lower limit of quantification (LLOQ) for values which are NQ.
- if more than 50%, but not all, of the concentrations are NQ, the gmean, CV, gmean ± SD, arithmetic mean and SD will be reported as not calculable (NC)
- if all the concentrations are NQ, the geometric mean and arithmetic mean will be reported as NQ and the CV, gmean ± SD and SD as NC

If the calculation of the geometric mean - SD results in a value less than the LLOQ, NQ will be displayed.

Pharmacokinetic parameters will be calculated using non-compartmental analysis. Plasma concentrations of AZD9668 below the LLOQ at the end of the curve will not be considered for pharmacokinetic analysis, however if concentrations below the LLOQ between measurable concentrations should be set to LLOQ/2. All pre-dose samples which are below LLOQ will be set to zero. The actual sampling time will be used for the analysis. The maximum plasma concentration (C_{max}) and the time to reach the maximum plasma concentration (t_{max}) will be determined by visual inspection of the plasma concentration-time profile. The area under the plasma concentration-time curve from time zero to 24 h after dosing $AUC_{(0-24)}$ and from zero to the time of the last quantifiable plasma concentration (t_{last}) $AUC_{(0-t)}$, will be calculated using the linear trapezoidal rule (linear/log interpolation). The rate constant of the slowest disposition phase (λ_z) will be calculated by log-linear regression of the terminal portion of the concentration-time profile where there were sufficient data (i.e. at least 3 time points and R^2 adjusted is greater than or equal to 0.9). If these criteria are not fulfilled, AUC, CL/F, $t_{1/2}$ and Vz/F will not be reported. The terminal half-life (t ½) will be derived from the equation ln(2)/ λ_z. The area under the plasma concentration-time curve from time zero to infinity (AUC) will be determined by using λ_z to extrapolate $AUC_{(0-t)}$ to infinity (provided less than or equal to

20% of the AUC is extrapolated). Total apparent drug clearance (CL/F) and the terminal volume of distribution (V_z/F) will be estimated by dividing the dose by AUC, and product of λ_z and AUC, respectively. The equivalent PK parameters determined at steady state will also be calculated.

All pharmacokinetic parameters (except t_{max}) will be summarised by the number of observations, geometric mean, coefficient of variation (CV, calculated as 100 x $\sqrt{}$ [exp (s^2)-1], where s is the standard deviation of the data on a log scale), arithmetic mean, standard deviation (SD) using untransformed data, median, minimum and maximum. t_{max} will be summarised using median, minimum and maximum and number of observations.

For the urine data the amount of drug recovered for each individual collection period and the total amount over the collection period will be calculated. In addition the percentage of drug recovered for each collection period (and the total percentage recovered over the collection period) and the overall CL_R will be calculated. Summary statistics will be the same as for the plasma PK parameters.

Plasma concentration profiles will be presented graphically as individual curves and geometric mean plots, by study day, on both linear and log concentration scales.

6.4.3 Pharmacodynamic measurement and variables

6.4.3.1 Induced sputum weight, neutrophils in sputum and other biomarkers

The ratio of the neutrophils in sputum (and other biomarkers and sputum weight) on active to placebo will be analysed. This will be performed using analysis of variance, modelling the data on a log scale and then back transforming the results, the baseline values will be included as covariates. If the data cannot be transformed then the data will be analysed using a non-parametric test (Wilcoxon rank sum) instead. The medians will be estimated using Hodge-Lehmann procedure.

6.4.4 Safety analysis

Examination of the following safety variables will address the primary objective to investigate the tolerability of 14 days' treatment with AZD9668. No formal hypotheses will be associated with these variables, although informal comparisons will be made between treatment groups to determine whether there appears to be any treatment-related effects: adverse events, vital signs, 12-lead ECG (overall evaluation), lung function, sputum bacteriology, safety lab data (clinical chemistry, haematology, urinalysis), physical examination and concomitant medication.

Adverse events will be analysed by means of descriptive statistics and qualitative analysis. AEs will be listed for each patient and summarised by System Organ Class and preferred term assigned to event by using MedDRA 10.0.

Concomitant medications will be classified according to the AZDD Drug Dictionary, the ATC system and the CPMP route of administration dictionary. All concomitant medications reported at entry and recorded during the study will be summarised and listed.

6.4.5 Secondary outcome and exploratory variables

See Section 6.4.3 Pharmacodynamic measurement and variables

BronkoTest© diary card data: PEF, symptom scores and sputum scores (including amount and colour), the markers of tissue degradation, and the inflammatory markers in plasma will be summarised, plotted and listed.

6.5 Determination of sample size

No formal statistical sample size calculation has been performed for this phase II trial. The number of patients included is based on feasibility.

6.6 Interim analyses

No interim analysis will be performed.

6.7 Data monitoring board

Not applicable in this study.

7. STUDY MANAGEMENT

7.1 Monitoring

Before first patient into the study, a representative of AstraZeneca will visit the investigational study site to:

- Determine the adequacy of the facilities
- Discuss with the investigator(s) (and other personnel involved with the study) their responsibilities with regard to protocol adherence, and the responsibilities of AstraZeneca or its representatives. This will be documented in a Clinical Study Agreement between AstraZeneca and the investigator
- Discuss the specific requirements of the genetic research with the investigator(s) (and other personnel involved with the study)

During the study, a monitor from AstraZeneca or company representing AstraZeneca will have regular contacts with the study site, including visits to:

- Provide information and support to the investigator(s)
- Confirm that facilities remain acceptable
- Confirm that the investigational team is adhering to the protocol, that data are being accurately recorded in the eCRFs/pCRFs, and that investigational product accountability checks are being performed

- Perform source data verification (a comparison of the data in the eCRFs/pCRFs with the patient's medical records at the hospital or practice, and other records relevant to the study). This will require direct access to all original records for each patient (eg, clinic charts).

- Perform source verification of the genetic consent of participating patients and ensure that the investigational team is adhering to the specific requirements of this genetic research.

The monitor or another AstraZeneca representative will be available between visits if the investigator(s) or other staff at the centre need information and advice.

7.2 Audits and inspections

Authorised representatives of AstraZeneca, a regulatory authority, an Ethics Committee may visit the centre to perform audits or inspections, including source data verification. The purpose of an AstraZeneca audit or inspection is to systematically and independently examine all study-related activities and documents to determine whether these activities were conducted, and data were recorded, analysed, and accurately reported according to the protocol, Good Clinical Practice (GCP), guidelines of the International Conference on Harmonisation (ICH), and any applicable regulatory requirements. The investigator should contact AstraZeneca immediately if contacted by a regulatory agency about an inspection at his or her centre.

7.3 Training of staff

The principal investigator will maintain a record of all individuals involved in the study (medical, nursing and other staff). He or she will ensure that appropriate training relevant to the study is given to all of these staff, and that any new information of relevance to the performance of this study is forwarded to the staff involved.

Before the first patient is entered into the study the investigational staff will have an opportunity to discuss the procedures associated with the collection of blood samples, extraction of DNA and genetic research with AstraZeneca personnel. The ethical considerations specific to genotyping and the importance of the informed consent process will be made clear. The requirements for the collections of the patients' samples will also be made clear.

7.4 Changes to the protocol

Study procedures will not be changed without the mutual agreement of the Co-ordinating Investigator and AstraZeneca.

If it is necessary for the study protocol to be amended, the amendment and/or a new version of the study protocol (Amended Protocol) must be notified to or approved by each Ethics Committee, and if applicable, also the local regulatory authority, before implementation. Local requirements must be followed.

If an administrative change is required, such a change must be must be notified to or approved by each Ethics Committee according to local requirements.

If a protocol amendment requires a change to a particular centre's Informed Consent Form, then AstraZeneca and the centre's Ethics Committee must be notified. Approval of the revised Informed Consent Form by AstraZeneca and by the Ethics Committee is required before the revised form is used.

AstraZeneca will distribute amendments and new versions of the protocol to each principal investigator(s), who in turn is responsible for the distribution of these documents to his or her Ethics Committee, and to the staff at his or her centre. The distribution of these documents to the regulatory authority will be handled according to local practice.

7.5 Study agreements

The principal investigator must comply with all the terms, conditions, and obligations of the Clinical Study Agreement for this study. In the event of any inconsistency between this Clinical Study Protocol and the Clinical Study Agreement, the Clinical Study Protocol shall prevail.

7.6 Study timetable and end of study

Before a patient's enrolment in the study and any study-related procedures are undertaken the following should be fulfilled:

- Signed Clinical Study Protocol and other agreements between AstraZeneca and the Principal Investigator/Study Site.
- Approval of the study by the Ethics Committee
- Approval of the study, if applicable, by the regulatory authority.

The end of study is defined as the date of database lock, which is the time point after which no patient will be exposed to study-related activities.

8. ETHICS

8.1 Ethics review

AstraZeneca will provide Ethics Committees and Principal Investigators with safety updates/reports according to local requirements.

The final study protocol, including the final version of the Informed Consent Form, must be approved or given a favourable opinion in writing by an Ethics Committee as appropriate. The investigator must submit written approval to AstraZeneca before he or she can enrol any patient into the study.

The Principal Investigator is responsible for informing the Ethics Committee of any amendment to the protocol in accordance with local requirements. In addition, the Ethics Committee must approve all advertising used to recruit patients for the study. The protocol must be re-approved by the Ethics Committee annually, as local regulations require.

Where there is a genetic research, approval must be obtained for this genetic research and the associated genetic informed consent from the Ethics Committee. It must be clearly stated in the approval that this genetic research is approved. The investigator must submit written approval to AstraZeneca before any patient participates in this genetic research.

8.2 Ethical conduct of the study

The study will be performed in accordance with ethical principles that have their origin in the Declaration of Helsinki and are consistent with ICH/Good Clinical Practice, applicable regulatory requirements and the AstraZeneca policy on Bioethics.

For studies including genetic analysis special precautions are taken as described in Section 4.9.

8.3 Informed consent

The principal investigator(s) at each centre will ensure that the patient is given full and adequate oral and written information about the nature, purpose, possible risk and benefit of the study. Patients must also be notified that they are free to discontinue from the study at any time. The patient should be given the opportunity to ask questions and allowed time to consider the information provided.

The patient's signed and dated informed consent must be obtained before conducting any procedure specifically for the study.

The principal investigator(s) must store the original, signed Informed Consent Form. A copy of the signed Informed Consent Form must be given to the patient.

The genetic research is optional and the patient may participate in the main study without participating in the genetic component. To participate in the genetic component of the study the patient must sign and date both the consent form for the main study (non-genetic components of the study) and the genetic component of the study. Copies of both signed and dated consent forms must be given to the patient and the original filed at the study centre. The principal investigator(s) is responsible for ensuring that consent is given freely and that the patient understands that they may freely discontinue the genetic aspect of the study at any time.

If modifications are made according to local requirements, the new version has to be approved by AstraZeneca.

8.4 Patient data protection

The Master Informed Consent Form will incorporate (or, in some cases, be accompanied by a separate document incorporating) wording that complies with relevant data protection and privacy legislation. Pursuant to this wording, patients will authorise the collection, use and disclosure of their study data by the Investigator and by those persons who need that information for the purposes of the study.

The Master Informed Consent Form will explain that study data will be stored in a computer database, maintaining confidentiality in accordance with national data legislation. All data computer processed by AstraZeneca will be identified by randomisation code.

The Master Informed Consent Form will also explain that for data verification purposes, authorised representatives of AstraZeneca, a regulatory authority, an Ethics Committee may require direct access to parts of the hospital or practice records relevant to the study, including patients' medical history.

Reference to participation in this genetic research should not be recorded into the patients' general medical records. All notes should be kept within the clinical study records.

Due to the exploratory nature of this genetic research, there will be no routine communication of results to patients. AstraZeneca will not provide individual genotype results to patients, any insurance company, any employer, their family members, general physician or any other third party, unless required to do so by law.

Extra precautions are taken to preserve confidentiality and prevent genetic data being linked to the identity of the patient, however, it must be recognised that there are exceptional circumstances where individuals may see both genetic data and a patient's personal identifier, for example in the case of a medical emergency, when AstraZeneca Physicians and investigators might know the patients' identity and might have access to the genetic data, or during regulatory audit where designated authorities must be permitted access to the relevant files.

9. PROCEDURES IN CASE OF EMERGENCY, OVERDOSE OR PREGNANCY

9.1 AstraZeneca emergency contact procedure

In the case of a medical emergency the Investigator may contact the Study Delivery Team Leader. If the Study Delivery Team Leader is not available, the Study Delivery Team Physician may be contacted at the AstraZeneca Research and Development site shown below.

Role in the study	Name	Address & telephone number
LSDT Leader/Monitor responsible for the Protocol at the CRO	[redacted]	[redacted] Phone: [redacted]
LSDT Physician responsible for the Protocol at the CRO	[redacted]	[redacted] Phone: [redacted]
The local AstraZeneca representative could be found in the 'Supplement Study Delivery Team Contacts in the Event of Emergency'		

9.2 Procedures in case of medical emergency

The principal investigator(s) is responsible for ensuring that procedures and expertise are available to handle medical emergencies during the study. **A medical emergency usually constitutes an SAE and should be reported as such, see Section 4.7.1.1.**

9.3 Procedures in case of overdose

- Use of study medication in doses in excess of that specified in the protocol should not be recorded in the eCRFs/pCRFs as an AE of 'Overdose' unless there are associated symptoms or signs.
- An overdose with associated SAEs should be recorded as the SAE diagnosis/symptoms on the relevant AE forms in the eCRFs/pCRFs.
- An overdose with associated non-serious AEs should be recorded as the AE diagnosis/symptoms on the relevant AE forms in the eCRFs/pCRFs. In addition, the overdose should be reported on the separate AZ "Clinical Study Overdose Report Form."
- An overdose without associated symptoms should not be recorded as an AE in the eCRFs/pCRFs. The overdose should be reported on the separate AZ "Clinical Study Overdose Report Form".

9.4 Procedures in case of pregnancy

In case of pregnancy the subject should be discontinued from the study and withdrawn from the treatment with investigational drug. Pregnancy itself is not regarded as an adverse event unless there is a suspicion that the investigational product under study may have interfered with the effectiveness of a contraceptive medication. However, the outcome of all pregnancies (spontaneous miscarriage, elective termination, normal birth or congenital abnormality) must be followed up, documented and reported to AstraZeneca using the Pregnancy Report and Pregnancy Outcome Report CRF modules even if the subject was discontinued from the study. The Pregnancy Report CRF is used to collect information before the outcome of the pregnancy is known; it should be completed as soon as possible after it has been identified that a patient received an AstraZeneca product during pregnancy and should be sent to AstraZeneca Clinical Drug Safety within 30 days.

All reports of congenital abnormalities/birth defects are SAEs. Spontaneous miscarriages should also be reported and handled as SAEs. Elective abortions without complications should not be handled as AEs. All outcomes of pregnancy must be recorded on the Pregnancy Outcome Report CRF and reported to AstraZeneca Clinical Drug Safety within AE or SAE timeframes if appropriate. Reports of normal outcomes should be sent to AstraZeneca Clinical Drug Safety within 30 days.

Regarding paternal exposure, pregnancy of the subject's partner is not considered to be an adverse event. However, all pregnancies following paternal exposure to study treatment should also be reported to AstraZeneca and the outcome (spontaneous miscarriage, elective termination, normal birth or congenital abnormality) followed up, even if the patient has discontinued from the study. The Pregnancy Outcome Report Form Parts I and II are used to collect and report pregnancy information to AstraZeneca if the partner of a male study participant becomes pregnant. The report of a pregnancy should be completed as soon as possible after it has been identified that a patient received an AstraZeneca product and should be sent to AstraZeneca Clinical Drug Safety within 30 days. All outcomes of pregnancy must be reported to AstraZeneca Clinical Drug Safety within AE or SAE timeframes if appropriate. Reports of normal outcomes should be sent to AstraZeneca Clinical Drug Safety within 30 days.

10. REFERENCES

GOLD 2005
Global Initiative for Chronic Obstructive Lung Disease. Global Strategy for the Diagnosis, Management, and Prevention of Chronic Obstructive Pulmonary Disease. Updated 2005. http://www.goldcopd.org/

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Halbert RJ, Natoli JL, Gano A Badamagarav E, Buist AS, Mannino DM. Global burden of COPD: systematic review and meta-analysis. Eur Respir J 2006; 28: 523-532.

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Rutten van Molken MPMH, Postma MJ, Joore MA, Van Genugten ML, Leidl R, Jager JC. Current and future medical costs of asthma and chronic obstructive pulmonary disease in The Netherlands. Respir Med 1999; 93: 779-787.

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National Heart, Lung, and Blood Institute. Morbidity & Mortality: Chartbook on Cardiovascular, Lung, and Blood Diseases. Bethesda, MD: US Department. of Health and Human Services, Public Health Service, National Institutes of Health, 1998. Available from: URL: http://www.nhlbi.nih.gov/resources/docs/02_chtbk.pdf.

Clinical Study Protocol: Appendix B

Drug Substance	AZD9668
Study Code	D0520C00002
Appendix Edition Number	1
Appendix Date	[redacted]

Appendix B
Additional Safety Information

FURTHER GUIDANCE ON THE DEFINITION OF A SERIOUS ADVERSE EVENT (SAE)

Life threatening

'Life-threatening' means that the subject was at immediate risk of death from the AE as it occurred or it is suspected that use or continued use of the product would result in the subject's death. 'Life-threatening' does not mean that had an AE occurred in a more severe form it might have caused death (eg, hepatitis that resolved without hepatic failure).

Hospitalisation

Out-patient treatment in an emergency room is not in itself a serious AE, although the reasons for it may be (eg, bronchospasm, laryngeal oedema). Hospital admissions and/or surgical operations planned before or during a study are not considered AEs if the illness or disease existed before the subject was enrolled in the study, provided that it did not deteriorate in an unexpected way during the study.

Important medical event or medical intervention

Medical and scientific judgement should be exercised in deciding whether a case is serious in situations where important medical events may not be immediately life-threatening or result in death, hospitalisation, disability or incapacity but may jeopardize the subject or may require medical intervention to prevent one or more outcomes listed in the definition of serious. These should usually be considered as serious.

Simply stopping the suspect drug does not mean that it is an important medical event; medical judgement must be used.

Example of such events is:

- Intensive treatment in an emergency room or at home for allergic bronchospasm

A GUIDE TO INTERPRETING THE CAUSALITY QUESTION

The following factors should be considered when deciding if there is a "reasonable possibility" that an AE may have been caused by the drug.

- Time Course. Exposure to suspect drug. Has the subject actually received the suspect drug? Did the AE occur in a reasonable temporal relationship to the administration of the suspect drug?

- Consistency with known drug profile. Was the AE consistent with the previous knowledge of the suspect drug (pharmacology and toxicology) or drugs of the same pharmacological class? OR could the AE be anticipated from its pharmacological properties?

- Dechallenge experience. Did the AE resolve or improve on stopping or reducing the dose of the suspect drug?

- No alternative cause. The AE cannot be reasonably explained by another aetiology such as the underlying disease, other drugs, other host or environmental factors.

- Rechallenge experience. Did the AE reoccur if the suspected drug was reintroduced after having been stopped? AstraZeneca would not normally recommend or support a rechallenge.

- Laboratory tests. A specific laboratory investigation (if performed) has confirmed the relationship?

A "reasonable possibility" could be considered to exist for an AE where one or more of these factors exist.

In contrast, there would not be a "reasonable possibility" of causality if none of the above criteria apply or where there is evidence of exposure and a reasonable time course but any dechallenge (if performed) is negative or ambiguous or there is another more likely cause of the AE.

In difficult cases, other factors could be considered such as:

- Is this a recognised feature of overdose of the drug?

- Is there a known mechanism?

Ambiguous cases should be considered as being a "reasonable possibility" of a causal relationship unless further evidence becomes available to refute this. Causal relationship in cases where the disease under study has deteriorated due to lack of effect should be classified as no reasonable possibility.

Clinical Study Protocol: Appendix C

Drug Substance	AZD9668
Study Code	D0520C00002
Appendix Edition Number	1
Appendix Date	[redacted]

Appendix C
WHO Risk Categories

Risk group	Shipping Requirement	Pathogen	Risk to individuals	Risk to the community	Examples of Pathogens and their Risk groups
1	Standard Diagnostic (IATA PI650)	A micro-organism that is unlikely to cause human disease.	NONE OR VERY LOW	NONE OR VERY LOW	Most bacteria, fungi and viruses
2	Standard Diagnostic (IATA PI650)	A pathogen that can cause human or animal disease but is unlikely to be a serious hazard to laboratory workers, the community, livestock or the environment. Laboratory exposures may cause serious infection, but effective treatment and preventive measures are available and the risk of spread of infection is limited.	MODERATE	LOW	Legionella pneumophila E. Coli 0157
3	Standard Diagnostic (IATA PI650)	A pathogen that usually causes serious human or animal disease but does not ordinarily spread from one infected individual to another. Effective treatment and preventive measures are available.	HIGH	LOW	HIV Hepatitis B Hepatitis C
4	High risk(IATA PI602)	A pathogen that usually causes serious human or animal disease and that can be readily transmitted from one individual to another, directly or indirectly. Effective treatment and preventive measures are not usually available.	HIGH	HIGH	Lassa Fever Ebola Virus

If a subject is being withdrawn due to a suspected infection in WHO risk categories 2, 3 and 4 no biological samples from this subject are allowed to be sent to the laboratory. Samples will be destroyed according to normal routines at the study site.

Clinical Study Protocol Amendment Number 01

Drug Substance	AZD9668
Study Code	D0520C00002
Date	[redacted]
Protocol Dated	[redacted]

A 2-week, randomised, double-blind, placebo-controlled, parallel group study to assess the tolerability and pharmacokinetics of orally administered AZD9668 in patients with COPD

This submission /document contains trade secrets and confidential commercial information, disclosure of which is prohibited without providing advance notice to AstraZeneca and opportunity to object.

Sponsor:

AstraZeneca AB, [redacted]

Centres affected by the Amendment:

This is a single centre study therefore only 1 centre is affected by the amendment.

The protocol for the study is to be amended as follows:

Section of protocol affected:

Section 3.3.2, Inclusion criteria No. 5, page 26

Previous text:

5. Post-bronchodilator FEV_1 30 - 80% predicted, FEV_1/FVC ratio < 70%

Revised text:

5. Post-bronchodilator FEV_1 30 - 80% predicted, FEV_1/FVC ratio < 70% (GOLD stage II to III, GOLD 2007)

Section of protocol affected:

Section 3.3.3, Exclusion criteria No. 12, page 27

Previous text:

6. QTc >450 ms or the presence of any arrhythmia in the ECG at Visit 1. (Premature ventricular or supraventricular ectopics up to 15/minute will be allowed as long as there are no other associated cardiac abnormalities).

Revised text:

12. (a) QTc >450 ms for males and >470 ms for females in the screening ECG

(b) The presence of any arrhythmia in the ECG at Visit 1. which in the opinion of the investigator may put the patient at risk or interfere with study assessments. (Premature ventricular or supraventricular ectopics up to 6/minute will be allowed at the investigators discretion, as long as there are no other associated cardiac abnormalities)

(c) Current or previous history of coronary artery disease, or congestive cardiac failure or any other clinically significant cardiac disease

Section of protocol affected:

Section 3.3.3, Exclusion criteria No. 15, page 28

Previous text:

15. History of excessive alcohol consumption or chronic alcohol induced disease

Revised text:

15. History of excessive alcohol consumption or chronic alcohol induced disease. Excessive alcohol consumption will be defined as the consumption >28 units/week in males or >21 units in females [1 unit is equivalent to half a pint (285 ml) of beer, 1 glass (125 ml) of wine or 1 measure (25 ml) of spirits].

Section of protocol affected:

3.3.5, Discontinuation of patients from treatment or assessment
3.3.5.1, Criteria for discontinuation, page 29

Previous text:

Patients may be discontinued from study treatment and assessments at any time at the discretion of the Investigator. The following could be specific reasons for discontinuing a patient from the study.

- Voluntary discontinuation by the patient who is at any time free to discontinue his/her participation in the study, without prejudice to further treatment

- Safety reasons as judged by the investigator and/or AstraZeneca

- Severe non-compliance to protocol as judged by the investigator and/or AstraZeneca

- Incorrect enrolment, i.e. the patient does not meet the required inclusion/exclusion criteria for the study

- Patient lost to follow-up

- Disease exacerbation, defined as an increase in respiratory symptoms requiring hospitalisation and/or a course of oral corticosteroids and/or antibiotics (either prescribed or self administered). Patients are advised to contact the investigator if they experience an increase in respiratory symptoms for 2 consecutive days, and where possible, prior to commencing on a course of corticosteroids or antibiotics.

- An ALT / AST level ≥3xULN confirmed by a repeat test, or any pattern of liver function test (LFT) abnormalities giving Investigator or AZ cause for concern

- Evidence of haemolysis as evidenced by the following:

  – Reticulocyte count of over 5%, **and**

  At least two of the following:

  – a drop in haemoglobin below 10.5 g/dl

  – decrease of 25% in haptoglobin. (The value post-dose to be compared with the average of two pre-dose values. The value should also be 15% below LLN).

  – increase of 25% in unconjugated bilirubin or lactate dehydrogenase (LDH). (The value post-dose to be compared with the average of two pre-dose values. The value should also be 15% above ULN)

- A platelet count of below 80 x 10^9/L confirmed by a repeat test (in the absence of obvious platelet clumping in a peripheral blood film).

Revised text:

Patients may be discontinued from study treatment and assessments at any time at the discretion of the Investigator. The following could be specific reasons for discontinuing a patient from the study.

- Voluntary discontinuation by the patient who is at any time free to discontinue his/her participation in the study, without prejudice to further treatment

- Safety reasons as judged by the investigator and/or AstraZeneca

- Severe non-compliance to protocol as judged by the investigator and/or AstraZeneca

- Incorrect enrolment, i.e. the patient does not meet the required inclusion/exclusion criteria for the study

- Patient lost to follow-up

- Disease exacerbation, defined as an increase in respiratory symptoms requiring hospitalisation and/or a course of oral corticosteroids and/or antibiotics (either prescribed or self administered). Patients are advised to contact the investigator if they experience an increase in respiratory symptoms for 2 consecutive days, and where possible, prior to commencing on a course of corticosteroids or antibiotics.

- An ALT / AST level ≥3xULN confirmed by a repeat test, or any pattern of liver function test (LFT) abnormalities giving Investigator or AZ cause for concern

- Evidence of haemolysis as evidenced by the following:

  – Reticulocyte count of over 5%, **and**

  At least two of the following:

  – a drop in haemoglobin below 10.5 g/dl

  – decrease of 25% in haptoglobin. (The value post-dose to be compared with the average of two pre-dose values. The value should also be 15% below LLN).

  – increase of 25% in unconjugated bilirubin or lactate dehydrogenase (LDH). (The value post-dose to be compared with the average of two pre-dose values. The value should also be 15% above ULN)

- A platelet count of below 80 x 10^9/L confirmed by a repeat test (in the absence of obvious platelet clumping in a peripheral blood film).

- A QTcF >500 ms on active treatment (QT interval corrected for heart rate by Fridericia method) or a QTcF prolongation >60 ms on average compared to baseline, confirmed by a repeat ECG in 30 minutes.

Section of protocol affected:

3.3.5, Discontinuation of patients from treatment or assessment
3.3.5.1, Criteria for discontinuation, Stopping criteria for the whole study, page 30

Previous text:

Stopping criteria for the whole study

- Two or more patients on active treatment are discontinued because of either an ALT/AST level ≥3 x ULN or any pattern of LFT abnormalities giving cause for concern

- Two or more patients on active treatment are discontinued because of evidence of haemolysis

- Two or more patients on active treatment are discontinued because of a platelet count of below 80 x 10^9/L

Revised text:

Stopping criteria for the whole study

- Two or more patients on active treatment are discontinued because of either an ALT/AST level ≥3 x ULN or any pattern of LFT abnormalities giving cause for concern

- Two or more patients on active treatment are discontinued because of evidence of haemolysis

- Two or more patients on active treatment are discontinued because of a platelet count of below 80 x 10^9/L

- A QTcF >500 ms on active treatment (QT interval corrected for heart rate by Fridericia method) or a QTcF prolongation >60 ms on average compared to baseline, confirmed by a repeat ECG in 30 minutes in 2 or more subjects on active treatment

Section of protocol affected:

Section 10, References, page 58

Added reference:

GOLD 2007
Global Initiative for Chronic Obstructive Lung Disease. Global Strategy for the Diagnosis, Management, and Prevention of Chronic Obstructive Pulmonary Disease. Updated 2007. http:www.goldcopd.org

Reason for Amendment:

The original protocol is amended as required by the comments of the Ethic Committee, Berlin, Germany, in order to follow the German Drug Law (*Arzneimittelgesetz*) §40, para 1, p.3, no.2, and exclude subjects with coronary heart disease and to add a cardiac stopping criteria. In addition, the term "excessive alcohol consumption" was to be defined more clearly.

Persons who initiated the Amendment:

The Study Delivery Team.

Clinical Study Protocol Amendment No 1 Appendix A	
Drug Substance	AZD9668
Study Code	D0520C00002
Edition Number	1
Date	[redacted]
Protocol Dated	[redacted]

Appendix A
Signatures

ASTRAZENECA SIGNATURE(S)

A 2-week, randomised, double-blind, placebo-controlled, parallel group study to assess the tolerability and pharmacokinetics of orally administered AZD9668 in patients with COPD

This Clinical Study Protocol and all Amendments to the CSP have been subjected to an internal AstraZeneca peer review.

I agree to the terms of this study protocol/amendment.

AstraZeneca Research and Development site representative

[redacted]

[redacted]

Medical Science Director
AstraZeneca R&D [redacted]

[redacted]

Date
(Day Month Year)

This document contains confidential information, which should not be copied, referred to, released or published without written approval from AstraZeneca. Investigators are cautioned that the information in this protocol may be subject to change and revision.

ASTRAZENECA SIGNATURE(S)

A 2-week, randomised, double-blind, placebo-controlled, parallel group study to assess the tolerability and pharmacokinetics of orally administered AZD9668 in patients with COPD

This Clinical Study Protocol and all Amendments to the CSP have been subjected to an internal AstraZeneca peer review.

I agree to the terms of this study protocol/amendment.

AstraZeneca Research and Development site representative

[redacted]

Date
(Day Month Year)

Study Delivery Team Leader
AstraZeneca R&D [redacted]

[redacted]

This document contains confidential information, which should not be copied, referred to, released or published without written approval from AstraZeneca. Investigators are cautioned that the information in this protocol may be subject to change and revision.

SIGNATURE OF PRINCIPAL INVESTIGATOR

A 2-week, randomised, double-blind, placebo-controlled, parallel group study to assess the tolerability and pharmacokinetics of orally administered AZD9668 in patients with COPD

This Clinical Study Protocol and all Amendments to the CSP have been subjected to an internal AstraZeneca peer review.

I agree to the terms of this amendment.

Centre No.: 01

Signature: [redacted]

Date
(Day Month Year)