AstraZeneca

Revised Clinical Study Protocol

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A Phase I, Open-Label, Multi-centre Study to Assess the Safety, Tolerability and Pharmacokinetics of Single and Multiple Oral Doses of AZD8330 in Patients With Advanced Malignancies

Sponsor: AstraZeneca AB, 151 85 Södertälje, Sweden.

AstraZeneca Research and Development site representative

Date

The following Amendment(s) and Administrative Changes are included in this amended protocol:

Amendment No.	Date of Amendment	Amendment No.	Date of Amendment
1		5	
2		6	
3		**Local Amendment No.**	**Date of local Amendment**
4			

Administrative change No.	Date of Administrative Change	Local Administrative change No.	Date of local Administrative Change

AstraZeneca

PROTOCOL SYNOPSIS

A Phase I, Open-Label, Multi-centre Study to Assess the Safety, Tolerability and Pharmacokinetics of Single and Multiple Oral Doses of AZD8330 in Patients With Advanced Malignancies

Principal Investigator

Co Investigators

Study centre(s) and number of patients planned

Multi-centre study (2 centres in US, 1 centre in Norway), with approximately 70 patients with advanced malignancies. The actual number recruited is dependent upon the number of dose escalations in Part A (Dose Escalation Phase).

Study period		**Phase of development**
Estimated date of first patient enrolled	April 2007	Phase I
Estimated date of last patient completed	August 2010*	

* End of study is defined as the point at which the last patient has received 6 months of treatment with AZD8330, or has withdrawn from the study (whichever is sooner).

Objectives

Primary

Primary Objective	Outcome variables
To assess the safety and tolerability of AZD8330 in patients with advanced malignancies.	Adverse Events Clinical chemistry (including BNP or NT-proBNP)[a], haematology, coagulation, and urinalysis Vital signs MUGA scans/echocardiography Electrocardiograms (ECGs) Ophthalmologic examination O_2 saturation

Secondary

Secondary Objectives	Outcome variables
To determine the pharmacokinetics (PK) of AZD8330 following both single and multiple oral dosing of AZD8330 in patients with advanced malignancies.	Derived pharmacokinetic parameters for AZD8330 will be produced following both a single and multiple oral dosing. This may include C_{max}, t_{max}, AUC, CL/F, and $t_{1/2}$
To investigate possible relationships between plasma AZD8330 concentrations/exposure and changes in safety and pharmacodynamic parameters.	Output from both graphical and/or appropriate PK/pharmacodynamic modelling techniques
To investigate the effect of AZD8330 treatment on pERK in PBMCs in patients with advanced malignancies.	Change in biomarker pERK in PBMCs

Exploratory	
Exploratory Objectives	**Outcome Variables**
To obtain a preliminary assessment of efficacy of AZD8330.	Objective Response Rate (ORR; based on RECIST) for patients with measurable disease Best Overall Response (BOR; based on RECIST) for patients with measurable disease Patients with non target disease will be assessed for complete response, stable disease or progression
To collect a blood sample (optional) for DNA extraction and storage to investigate whether variability in the AZD8330 PK, safety, efficacy or pharmacodynamic results could be explained by differences in the patient's genotype.	Correlation of host polymorphisms with variation in PK, safety or response parameters observed in patients treated with AZD8330

[a] BNP or NT-proBNP are permitted in the protocol, however these shall be collectively referred to as BNP for the remainder of the protocol.

Study design

The study is in 1 part, as briefly described below:

Part A: Dose escalation phase

- Part A aims to define the maximum tolerated dose (MTD) of AZD8330, and in the process collect data to allow evaluation of safety, tolerability and pharmacokinetics (PK) during the escalation phase. Six to nine patients will be enrolled per cohort.

The design as described will provide an initial estimate for the maximum tolerated dose (MTD) following once daily (OD) or twice daily (BD) dosing in Part A of the study.

Target patient population

Male and female patients aged 18 years or older who have a cancer that is refractory to standard therapies or for which no standard therapies exist. Inclusion is irrespective of stage of disease.

Investigational product, dosage and mode of administration

AstraZeneca Investigational Products section will supply AZD8330 open labelled clinical study material.

AZD8330 tablets will be supplied in 0.5 mg, 0.75 mg, 1.5 mg, and 5 mg strengths in high-density polyethylene (HDPE) bottles; each bottle will contain sufficient tablets for each treatment period, as appropriate.

Duration of treatment

Patients may continue to receive AZD8330 until disease progression and as long as they are continuing to derive benefit from treatment.

Patients who continue to receive treatment beyond the defined end of the study, in accordance with Section 7.6 (Study timetable and end of study) will be followed up according to the investigational site standard of care and investigator judgement. Investigators must continue to report all Serious Adverse Events to AstraZeneca Patient Safety Department until 30 days after study treatment is discontinued in accordance with Section 4.5.1.2 (Recording of adverse events) and 4.5.1.3 (Reporting of serious adverse events).

Statistical methods

No formal statistical analysis will be performed on the data from this study. Both the AZD8330 concentration-time data and derived pharmacokinetic parameters will be summarised using descriptive statistics. Pharmacodynamic data will be summarised descriptively using confidence intervals if sufficient data are available. Safety data will be listed and summarised. Graphical presentations of the data will be produced to aid interpretation.

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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
AE	Adverse event (see definition in Section 4.5.1.1)
ALT	Alanine aminotransferase
ANOVA	Analysis of variance
APTT	Activated Partial Thromboplastin Time
ASCO	American Society of Clinical Oncology
AST	Aspartate aminotransferase
AUC	Area under the plasma concentration-time curve from zero to infinity
AUC(0-t)	Area under the plasma concentration-time curve from zero to time of the last quantifiable concentration
AUCτ	Area under the plasma concentration-time curve at steady-state for dosing interval
AUC(0-24)	Area under the plasma concentration-time curve from zero to 24 hours post dose
BCRP	Breast Cancer Resistance Protein (ATP-binding cassette, sub-family G (WHITE), member 2)
BD	Twice daily dosing
BNP	b-type natriuretic peptide
BOR	Best Overall Response
BP	Blood pressure
B-RAF	v-raf murine sarcoma viral oncogene homolog B1
pCRF	Paper Case Report Form
CI	Confidence interval
Cmax	Maximum plasma concentration
CL/F	Total apparent drug clearance
CNS	Central Nervous System
CR	Complete response
C-RAF	v-raf-1 murine leukemia viral oncogene homolog 1
CRO	Contract Research Organisation
CSA	Clinical Study Agreement

Abbreviation or special term	Explanation
CT	Computed Tomography
CTCAE	Common Terminology Criteria Adverse Event
CTC	Common terminology criteria
CYP	Cytochrome P
DBL	Database Lock
DOB	Date of birth
DLT	Dose Limiting Toxicity
E number	Enrollment number
ECG	Electrocardiogram
EF	Ejection fraction
ERK	Extracellular signal-regulated kinase
Ethics Committee	Synonymous to Institutional Review Board and Independent Ethics Committee
FDA	Food and Drug Administration (United States department of health and human services)
gmean	Geometric Mean
glsmean	Geometric Least Squares Mean
GCP	Good Clinical Practice
Fl oz	Fluid Ounce
GI	Gastrointestinal
GMP	Good manufacturing practice
HDPE	High density polyethylene
HED	Human Equivalent Dose
H-RAS	v-Ha-ras Harvey rat sarcoma viral oncogene homolog
HRCT	High Resolution CT
IB	Investigator Brochure
ICH	International Conference on Harmonisation
IHC	Immunohistochemistry
IP	Investigational Product
IPS	Investigational Product Supplies
ITT	Intention to treat

Abbreviation or special term	Explanation
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.
Ki-67	IHC marker of Cell Proliferation
K-RAS	v-Ki-ras2 Kirsten rat sarcoma viral oncogene homolog
MAP	Mitogen Activated Protein
MAPKK	Mitogen-Acitvated Protein Kinase Kinase
MEDRA	Medical dictionary for regulatory activities
MEK	Mitogen-Activated Protein Kinase Kinase (also known as MAPKK)
mg	Milligrams
MRI	Magnetic Resonance Imaging
MUGA	Multiple gated acquisition scan
MTD	Maximum Tolerated Dose
NA	Not applicable
N-RAS	neuroblastoma RAS viral (v-ras) oncogene homolog
NSTD	Non Serious Toxicity Dose
nM	Nanomolar
NT-proBNP	N-terminal prohormone b-type natriuretic peptide
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 4.5.1.1).
OD	Once daily dosing
ORR	Objective Response Rate
PD	Progressive Disease
PBMC	Peripheral blood mononuclear cell (leucocyte ghosts)
PFT	Pulmonary function test
pERK	Phosphorylated Extracellular signal-regulated kinase
PGP	P glycoprotein
PK	Pharmacokinetics
PR	Partial response
QWBA	Qualitative Whole Body Autoradiography
RECIST	Response evaluation criteria in solid tumours
SAE	Serious adverse event (see definition in Section 4.5.1.1).

Abbreviation or special term	Explanation
SAP	Statistical Analysis Plan
SD	Stable Disease
SOC	System Organ Class
STD	Severely Toxic Dose
TPA	12-0-Tetradecanoyl phorbol-13-acetate
tmax	Time to reach maximum plasma concentration
t1/2	Half life
Tx	Treatment
ULN	Upper limit of normal
UV	Ultra Violet
WHO	World Health Organisation

1. INTRODUCTION

1.1 Background

1.1.1 Mitogen activated protein kinase (MEK)

Human cancers often arise as a result of mutations in cellular signalling pathways that coordinate and regulate cell proliferation and survival. The intracellular Ras regulated RAF/MEK/ERK protein kinase signal cascade is a key pathway involved in cellular proliferation and there is a strong link between deregulation of this pathway and uncontrolled cell proliferation and survival (Chow et al 2005). The RAF/MEK/ERK kinase cascade holds a central coordinating role in mediating signal transduction from extra cellular growth factors, cytokines and proto-oncogenes. Some tumours contain mutations that result in the continuous activation of the pathway due to continuous production of growth factors, or express mutated or increased levels of growth factor receptors. Within this central cascade, mitogen activated protein kinase kinase (MEK) has been identified as an attractive therapeutic target because the only known substrates for MEK phosphorylation are the mitogen activated protein (MAP) kinases, extracellular signal regulated kinase ERK1 and ERK2.

It is anticipated that inhibition of MEK activity should inhibit transduction of the mitogenic signals from multiple pathways, resulting in an effect on tumour proliferation, differentiation and survival. Given the widespread distribution of constitutive ERK activation, a MEK inhibitor could be anticipated to have a broad spectrum of clinical utility in both solid tumours and haematological malignancies.

AZD8330 is a potent, selective, uncompetitive inhibitor of MEK, in-licensed for development by AstraZeneca Pharmaceuticals from Array BioPharma.

AZD8330 pre-clinical information is summarised in Section 1.1.2 of this protocol, and further information on pre-clinical toxicology and pharmacology can be found in the IB.

1.1.2 Pre-clinical experience with AZD8330

In vitro studies with enzyme assay have demonstrated that AZD8330 inhibits the activity of isolated MEK with IC_{50} values of 8 nM. In contrast to its activity against MEK, AZD8330 was inactive against a panel of 35 other kinases even when tested at a concentration of 10 µM.

There was no clear evidence of unique human metabolites. AZD8330 showed no inhibition of the main CYP enzymes with the exception of 2C9 and 2C19. Given the likely pharmacologically relevant plasma concentrations (predicted to be less than 1µM (approx. 461ng/mL)) it is considered unlikely that AZD8330 will cause clinically relevant inhibition of the CYP pathways. AZD8330 was an inducer of CYP3A4 in human hepatocytes with induction up to 16% of a positive control inducer (rifampicin) at 0.01µM, up to 55% at 0.1µM and up to 97% at 1µM. The likely impact of this induction will

need to be assessed once pharmacologically active concentrations are known in the clinic. AZD8330 caused minimal induction of CYP1A and induction experiments on CYP2C9 were inconclusive. AZD8330 inhibited MEK dependent phosphorylation of ERK1/2 in cellular assays with IC_{50} values of 0.4 nM. In addition, it inhibited cell viability in a range of tumour cell lines for example in colorectal HT-29 cells with an IC_{50} of 3.8 nM.

Treatment of mice and rats bearing a variety of xenografted tumours including lung and colorectal carcinomas with AZD8330 resulted in dose-dependent inhibition of tumour growth at well-tolerated doses. For example treatment of mice bearing Calu-6 xenograft lung tumours with 1 mg/kg twice daily (BD) resulted in 96.4% tumour growth inhibition. In studies conducted in rats bearing the Calu-6 tumour, doses as low as 0.4 mg/kg once daily are highly effective. *In vivo* pharmacodynamic inhibition of the relevant target was demonstrated in mice bearing HT-29 colorectal xenografted tumours. A single 10 mg/kg dose of AZD8330 resulted in a greater than 90% inhibition of ERK1/2 phosphorylation at both 1 and 4 hours post dose.

The excretion balance studies suggested that absorption in rat and dog was high. In rat and dog there was a greater than proportional increase in exposure with increasing dose which may suggest saturation of a transporter or metabolic enzyme with increasing dose. There was no/minimal accumulation of AZD8330 on multiple dosing in rat whereas 1.4 to 4-fold accumulation occurred in dog. In vitro protein binding values were 90.7% rat, 82.0% dog and 91.4% human. In the rat qualitative whole body autoradiography (QWBA) study radioactivity was widely distributed with minimal penetration into the CNS and no evidence of protracted binding to melanin-containing tissues. In rat and dog, drug-related material was primarily associated with the plasma component of blood. AZD8330 is a substrate but not an inhibitor of PGP and BCRP.

Soft tissue mineralization (most notably in glandular stomach, kidney, heart, lungs or aorta) was seen in rats following single oral doses at high dose levels, or following daily oral dosing for 1 month. Tissue mineralization was also seen in one female cynomolgus monkey (stomach, ovary) dosed with AZD8330 at escalating dose levels over a 21 day period, and in dogs (heart papillary muscle, pyloric stomach, ovary) following dosing for 3 or 7 days. In dogs dosed with AZD8330 for 1 month, minimal pyloric stomach mucosal mineralization was seen in small numbers of animals, usually in association with lamina propria histiocytic foci. In rats, dogs and primates, the tissue mineralization was associated with changes in plasma calcium, inorganic phosphate, calcium:phosphate ratio and/or albumin, and, in rats, with changes in urinary phosphate and calcium. Soft tissue mineralization, with associated changes in plasma calcium and/or inorganic phosphate, has been seen in mice and rats dosed with other MEK inhibitors, including AZD6244 and PD0325901. To date no changes in plasma calcium or phosphate have been reported in Phase I clinical trials with these agents.

Dosing of AZD8330 was associated with gastrointestinal tract changes in rats, cynomolgus monkeys and dogs. In rats, single oral dosing at doses of 500 μmol/kg and above caused enteropathy, which was not apparent following dosing for 1 month at lower doses. In cynomolgus monkeys, loose/liquid faeces and signs of dehydration were seen but with no

associated histopathological changes in the gastrointestinal tract. In dogs, oral dosing for 3 or 7 days was associated with vomiting, red coloured faeces, loss of skin tone, lack of appetite and/or body weight loss, with histopathological changes of gastrointestinal tract inflammation and gastric epithelial erosions. In the one-month dog study, gastrointestinal tract inflammatory changes were seen in high dose dogs, with some stomach inflammation in animals at lower doses.

Skin lesions, comprising multifocal bacterial colonies, crusting, ulceration, scab formation, dermatitis, and dermal or epidermal micro-abscesses were seen in rats following dosing with AZD8330 for 1 month. These changes were seen at all dose levels. Similar changes were not apparent following dosing of cynomolgus monkeys over a 21-day period, or in dogs dosed for up to 1 month.

In dogs, oral dosing for 1 month was associated with clinical pathology changes indicative of blood loss (decreases in red blood cell indices, reticulocytes) and inflammation (increased neutrophils, monocytes), and with a prolongation of APTT and increased fibrinogen. Haematology changes suggestive of blood loss and inflammation were also seen in cynomolgus monkeys dosed with AZD8330. In rats, dosing with AZD8330 for 1 month produced a slight reduction in haemoglobin at the high dose level, and clinical pathology changes indicative of inflammation at all dose levels. However, no changes in APTT or fibrinogen were apparent.

In male dogs, oral dosing with AZD8330 for 1 month was associated with degeneration and atrophy of the prostate and testes (with associated changes in the epididymis). Similar changes were not apparent in male rats dosed for 1 month with AZD8330, or in rats, mice or cynomolgus monkeys dosed with AZD6244. In female rats, oral dosing with AZD8330 for 1 month was associated with corpora luteal necrosis. Similar changes were not apparent in female dogs dosed for 1 month with AZD8330, or in rats, mice or cynomolgus monkeys dosed with AZD6244.

AZD8330 absorbs in the UV range for phototoxicity, showed clearly enhanced cytotoxicity in the presence of UV light in an *in vitro* phototoxicity test and is detected in both the skin and uveal tract in a rat. These data indicate a potential phototoxicity risk for AZD8330.

AZD8330 was not mutagenic *in vitro* in the Ames test or in the mouse lymphoma assay, and was not clastogenic in an *in vivo* bone marrow micronucleus assay in the mouse.

1.1.3 Clinical experience with AZD8330

D1536C00001 is the first clinical study to be conducted in the AZD8330 clinical development program. Based upon emerging safety data, the study has established a maximum tolerated dose (MTD) of 20mg once daily (OD) dosing. The MTD was defined as a result of 5 adverse events, verbatim investigator reported terms: “hallucination” and “mental status changes” at a frequency of 4 out of 9 patients at 40mg and 1 out of 3 patients at 60mg OD dosing. These adverse events did not meet the key DLT criteria (section 3.1.4.2), but met the following:

- Any toxicity that, in the opinion of the investigator, is sufficiently clinically significant to be considered a DLT.

Apart from the DLT criteria all other adverse events observed have been generally well tolerated.

AZD8330 was well distributed in the tissues clinically, as measured by Vss/F in this study. Pre-clinically there was minimal accumulation observed in the CNS (section 1.1.2). Patients received a single dose of AZD8330 on Day 1, followed by a washout period and OD continuous dosing from Day 8 onwards. The elimination half-life was approximately 15h, therefore steady state was achieved by Day 15 and observed accumulation of AZD8330 was minimal. The adverse events that affected mental status occurred, on average, 32 days (range 15 to 47 days) post- the first dose of AZD8330, no relationship was observed between exposure, as measured by Cmax and AUC, on Day 1 and 15, and the adverse events defined as mental status changes. Maximal AZD8330 plasma concentrations were observed approximately 0.5 to 1h post-dose and were also not temporally related to the adverse events. There is no known mechanistic rationale to explain these adverse events defining the MTD.

1.1.4 Clinical experience with other MEK inhibitors

Preliminary data reported from early clinical trials of other MEK inhibitors indicate a number of frequently reported adverse events including skin rash (acneiform), diarrhoea, fatigue, nausea, vomiting and peripheral oedema (Adjei A et al 2006, LoRusso P et al 2005). Dyspnoea / hypoxia and increased liver transaminases have also been reported with AZD6244 (Adjei A et al 2006), and alterations in visual function such as blurred vision have been seen in trials with different MEK inhibitors (Adjei A et al 2006, LoRusso P et al 2005). These agents have been associated with prolonged stable disease or partial responses in some patients in Phase I.

1.2 Rationale

This study is designed to evaluate for the first time the safety, tolerability and pharmacokinetic (PK) profile of AZD8330, when administered as a single or twice daily dose and continuous once or twice daily dosing by the oral route, to patients with advanced malignancies. The design of this study will provide data on the pharmacodynamic activity of AZD8330 as demonstrated by the effect on pERK in PBMCs post-dose compared to pre-dose. This will allow an assessment of the biological activity of the drug prior to commencement of further studies. Further rationale regarding the study design and dose selection is provided in Section 3.2.1 of this protocol.

2. STUDY OBJECTIVES

2.1 Primary objective

Primary Objective	Outcome variables
To assess the safety and tolerability of AZD8330 in patients with advanced malignancies.	Adverse Events Clinical chemistry (including BNP or NT-proBNP)[a], haematology, coagulation, and urinalysis Vital signs MUGA scans/echocardiography Electrocardiograms (ECGs) Ophthalmologic examination O_2 saturation

2.2 Secondary objectives

Secondary Objectives	Outcome variables
To determine the pharmacokinetics (PK) of AZD8330 following both single and multiple oral dosing of AZD8330 in patients with advanced malignancies.	Derived pharmacokinetic parameters for AZD8330 will be produced following both a single and multiple oral dosing. This may include C_{max}, t_{max}, AUC, CL/F, and $t_{1/2}$
To investigate possible relationships between plasma AZD8330 concentrations/exposure and changes in safety and pharmacodynamic parameters.	Output from both graphical and/or appropriate PK/pharmacodynamic modelling techniques
To investigate the effect of AZD8330 treatment on pERK in PBMCs in patients with advanced malignancies.	Change in biomarker pERK in PBMCs

2.3 Exploratory objectives

Exploratory objectives	Outcome variables
To obtain a preliminary assessment of efficacy of AZD8330	Objective Response Rate (ORR; based on RECIST) for patients with measurable disease Best Overall Response (BOR; based on RECIST) for patients with measurable disease Patients with non target disease will be assessed for complete response, stable disease or progression

To collect a blood sample (optional) for DNA extraction and storage to investigate whether variability in the AZD8330 PK, safety, efficacy or pharmacodynamic results could be explained by differences in the patient's genotype	Correlation of host polymorphisms with variation in PK, safety or response parameters observed in patients treated with AZD8330

[a] BNP or NT-proBNP are permitted in the protocol, however these shall be collectively referred to as BNP for the remainder of the protocol.

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.

3.1.1 Study design

This is a Phase I, open-label, multi-centre study to assess the safety, tolerability and pharmacokinetics of an oral formulation of AZD8330 in patients with advanced malignancies who have failed standard therapy, or for which no standard therapies exist. Inclusion is irrespective of stage of disease. The study is expected to recruit up to a maximum of approximately 70 patients from approximately 3 centres within the following countries: United States, and Norway.

The actual numbers recruited is dependent upon the number of dose escalations in Part A.

Reasons for patient withdrawal will be given due consideration when enrolling patients.

The study is in 1 part, as briefly described below:

Part A: Dose escalation phase

- Part A aims to define the maximum tolerated dose (MTD) of the oral formulation, and in the process collect data to evaluate the safety, tolerability and PK profile during the escalation phase. 6 to 9 patients will be enrolled per cohort.

The design as described will provide an initial estimate for the maximum tolerated dose (MTD) following once daily (OD) or twice daily (BD) dosing in Part A.

3.1.2 Study assessments

Following informed consent, patients will be screened for eligibility to receive study treatment. Screening assessments are described in more detail in Section 4.2, and include evaluation of clinical chemistry, haematology, coagulation, urinalysis, a physical examination, pregnancy test ((urine or serum) female pre-menopausal patients), vital signs, chest x-ray and ECGs. A full ophthalmological examination is required prior to the first dosing day. All screening assessments must be conducted within 2 weeks prior to receiving first study

treatment. Tumour assessment using RECIST (if measurable) or other (if non-measurable) criteria as appropriate, and MUGA/echocardiogram for assessment of cardiac function, do not need to be repeated if conducted within 4 weeks prior to starting treatment if already performed as part of standard clinical care.

Patients will attend a clinic visit on day 1 pre-first dose for assessment of AEs, clinical chemistry (including BNP), haematology, coagulation, urinalysis, O_2 saturation measurement, a pregnancy test ((urine or serum) female pre-menopausal patients), ECG, physical examination, and vital signs including weight.

Patients will have blood samples taken for pharmacokinetic and pharmacodynamic assessment as specified in the Study Plan (Table 1).

Patients will subsequently attend clinic visits at weeks 1, 2, 3 and 4, 5 (Part A only), and 8, then every 4 weeks thereafter, for assessment of AEs, clinical chemistry (including BNP and Troponin T (or Troponin I)), haematology, coagulation, urinalysis, pulse oximetry and vital signs.

Patients will also undergo physical exam at weeks 1, 2, 3 and 4, 5 (Part A only), and 8, then every 8 weeks thereafter.

Patients will have ECGs performed in triplicate prior to first dose, and at 1 hour 30 minutes and 4 hours after dosing on Day 1 and Day 15 in Part A, and at any cardiac event with symptoms that may be due to cardiac ischaemia or arrhythmia (such as chest pain or palpitations). The timing of the ECGs may be amended according to the emerging PK profile, however the total number of ECGs will not change.

O_2 saturation will be measured by pulse oximetry at every vital sign visit and if there is a clinically significant drop in O_2 saturation, this should be followed up with full pulmonary function tests (PFTs) and high resolution CT (HRCT) scan of the chest. ECGs, chest X- Ray, and O_2 saturation will be performed on patients in cases of dyspnoea or pulmonary oedema in addition to other assessments as per local standard practice.

A MUGA scan and/or echocardiogram will be performed at screening, week 3 and week 5 (+/- 1 week). Further scans are required should signs of congestive heart failure occur after this time point.

Pregnancy tests (for female pre-menopausal women) will be repeated every 8 weeks whilst on study.

Tumour assessments will be performed preferably using contrast enhanced CT or MRI (see Appendix C for assessment methods) every 8 weeks (relative to date of first dose with AZD8330) with a +/-7 day visit window.

A complete ophthalmologic examination must be performed 2 weeks prior to first dose (Day 1), and at week 5 (+/- 1 week). If a patient receiving AZD8330 experiences an AE of visual

disturbance (for example blurring of vision) a complete ophthalmologic examination including slit-lamp examination must be performed.

Following withdrawal from AZD8330 for any reason, patients will have clinical chemistry/haematology, coagulation, urinalysis, physical examination, vital signs, ECG, and AE assessments.

Following withdrawal from randomised treatment for any reason, patients may receive any subsequent therapy for their disease at the discretion of the investigator. Details of such treatment are to be recorded in the pCRF.

All patients will be followed until withdrawal of consent or the end of the study (the point at which the last patient recruited has received 6 months of treatment with AZD8330 or withdrawn from the study, whichever is sooner). Where possible, adverse events will be followed to resolution.

Patients who continue to receive treatment beyond the defined end of the study, in accordance with Section 7.6 (Study timetable and end of study) will be followed up according to the investigational site standard of care and investigator judgement. Investigators must continue to report all Serious Adverse Events to AstraZeneca Patient Safety Department until 30 days after study treatment is discontinued in accordance with Section 4.5.1.2 (Recording of adverse events) and 4.5.1.3 (Reporting of serious adverse events).

For the first cycle in Part A, patients should avoid changes to or the addition of all concomitant medications, in particular any that are likely to affect the metabolism of AZD8330 (e.g. inhibitors/inducers of CYP2C9/19), unless considered clinically essential for management of concurrent conditions (see Section 3.6).

3.1.3 Duration of treatment

In Part A, patients may continue to receive AZD8330 until disease progression and as long as they are continuing to derive benefit from treatment.

3.1.4 Part A: Dose Escalation Phase

Six patients will be recruited to the first cohort to receive a single 0.5 mg dose of AZD8330 on Day 1, followed by continuous 0.5 mg once daily dosing from Day 8. The starting dose administered was calculated based on preclinical toxicology results using the FDA starting dose calculation for oncology compounds. If this dose is tolerated then a further six patients will be recruited to a higher dose and dose escalation will continue until a non-tolerated dose is reached.

Replacement patients may be enrolled until the minimum required number of evaluable patients are available for assessments. Reasons for patient withdrawal will be given due consideration when enrolling replacement patients.

No more than 3 patients will start dosing in the first seven days of a cohort opening, after which a further 3 patients may start dosing. See Section 3.2.1 for further rationale on the starting dose and study design.

3.1.4.1 Dose Escalation Criteria to Maximum Tolerated Dose

Prior to each dose escalation in Part A, the Safety Review Committee (see Section 6.8) will meet to review the available safety and PK data. The dose escalation decision will be formally minuted and the minutes distributed to each site. Cohort dose escalation/de-escalation decisions in Part A, will be taken by the Safety Review Committee. The dose escalation/de-escalation decisions will be performed following review of available clinical data, the observed number of DLTs (see Section 3.1.4.2 for DLT definitions), PK data (C_{max} and AUC), and all safety assessments up to Day 35. Patients must have had sufficient safety evaluations performed during the 35 day dosing period, at the discretion of the Safety Review Committee. All dose escalation decisions by the Safety Review Committee will take into account the anticipated magnitude of plasma AZD8330 exposure in addition to the safety profile.

The decision criteria for dose escalation in any group of 6 to 9 patients are:

- For each group a Safety Review Committee will be held as soon as possible after 6 patients complete at least 35 days of treatment[1].
- The Safety Review Committee will define the dose for that group as a tolerated dose or a non-tolerated dose.
  - A non-tolerated dose is one where >25% of the patients experience a DLT. That is a dose where a dose limiting toxicity (DLT) is observed in ≥2/6, ≥2/7, ≥3/8, or ≥3/9 patients.
  - A tolerated dose is one where ≤25% of the patients experience a DLT. That is, a dose where a DLT is observed in ≤1/6, ≤1/7, ≤ 2/8, or ≤2/9.
- If the dose is defined as tolerated, an increased dose will be investigated in another group of 6 to 9 patients.
- Recruitment to a group will be terminated if the number of DLTs observed in a group meets the above criteria for a non-tolerated dose before recruitment to that group is completed.
- No patients will be recruited to a cohort after the Safety Review Committee for that cohort has met and defined the dose as tolerated or non-tolerated.
- In the event that DLTs are observed in a group within 35 days of commencing treatment such that a dose initially considered tolerated by the Safety Review Committee becomes non-tolerated and dose escalation has already occurred:

  - Recruitment to the escalated dose group will cease, and
  - Patients already recruited to the escalated dose group will either continue at the higher dose or reduce to some lower dose at the discretion of the Safety Review Committee.

- Doses will be escalated by no more than 100% until the persistence of a drug related CTC Grade 2 toxicity, despite optimal therapy (apart from rash-related / dermatological toxicities, where a CTC Grade 3 threshold will apply), after which further dose escalations will be no more than 50%. The Safety Review Committee will consider all clinical findings in the completed cohort when making dose escalation decisions.

To investigate the magnitude of possible non-linearity in systemic drug exposure, the option exists for subsequent cohorts (ie cohort 2 and onwards) to receive a lower single dose on Day 1 compared to Day 8.

When a non-tolerated dose (NTD) is defined, dose escalation will be stopped. An interim dose (ie, one between the non-tolerated dose and the last dose tested before non-tolerated dose) may be assessed to determine the MTD and the Safety Review Committee will decide this upon review of the data. The maximum tolerated dose (MTD) will be defined as the last dose assessed below the non-tolerated dose.

If DLT is observed in ≥ 2 patients in the first dose cohort, a second cohort of 6 patients will be enrolled to receive 50% of the first dose. De-escalation in this way may continue until MTD is defined.

There will be no intra-patient dose escalation of AZD8330 during the continuous once daily dose period in this study. If patient experiences an AZD8330 related toxicity, their dose may be reduced or withheld. Re-escalation will not be permitted. See Section 3.2.1 for further rationale around the dose escalation and study design.

3.1.4.2 Definition of a Dose Limiting Toxicity

A dose limiting toxicity is toxicity considered related to treatment with AZD8330, occurring in the first 35 days of treatment that is:

- Haematological
  - any CTC Grade 4 toxicity (CTC Grade 4 neutropaenia has to be present for >7 consecutive days to constitute a DLT)
  - ≥ CTC Grade 3 neutropaenia with fever. Fever is defined as a temperature of >37.5°C orally, 37.2°C axillary or 38°C rectally measured

- ≥ CTC Grade 3 thrombocytopaenia associated with bleeding (excluding patients receiving therapeutic systemic anticoagulation)

- Any non-haematological CTC Grade 3 or 4 toxicity despite adequate supportive care that is not clearly related to underlying disease.

- Any continuous dose interruption for >2 weeks for any toxicity considered by the Investigator or AstraZeneca to be possibly related to AZD8330.

- Any toxicity that, in the opinion of the investigator, is sufficiently clinically significant to be considered a DLT.

3.1.4.3 Management of patients with an Adverse Event

Treatment with AZD8330 should be withheld if one of the following toxicities is observed despite supportive care:

- Intolerable adverse event

- Any adverse events ≥CTC Grade 3

Treatment with AZD8330 should not be restarted until the toxicity improves to CTC Grade 1 or baseline. Treatment with AZD8330 may be resumed at the original dose or at a permanently reduced dose (to be agreed with AstraZeneca responsible medical officer).

If a patient experiences recurrence of an adverse event, study medication should again be withheld until the event improves to CTC Grade 1 or baseline. Upon recovery, treatment with AZD8330 may resume at a permanently reduced dose (to be agreed). Note: If the dose was not reduced following the first occurrence of the toxicity, the dose must be reduced upon a second occurrence (ie, treatment may only resume at the previous dose after the first occurrence of the toxicity). Dose re-escalation is not permitted in this study.

If an adverse event CTC Grade ≥3 recurs after a second dose reduction, the patients must permanently discontinue study treatment. All dose reductions will be recorded in the appropriate CRF.

In the event of a dose delay/reduction, patients should continue to follow the assessments schedule as described in Section 3.1.2, relative to baseline, with the exception of PK and peripheral blood mononuclear cell (PBMC) sampling. In the event of a missed dose occurring prior to completion of PK sampling in the study, investigators should contact AstraZeneca for guidance regarding rescheduling these assessments.

3.1.5 Part B: Safety Expansion Phase (Not applicable)

No longer applicable.

Figure 1 Study flow chart

Part A – Dose Escalation Phase

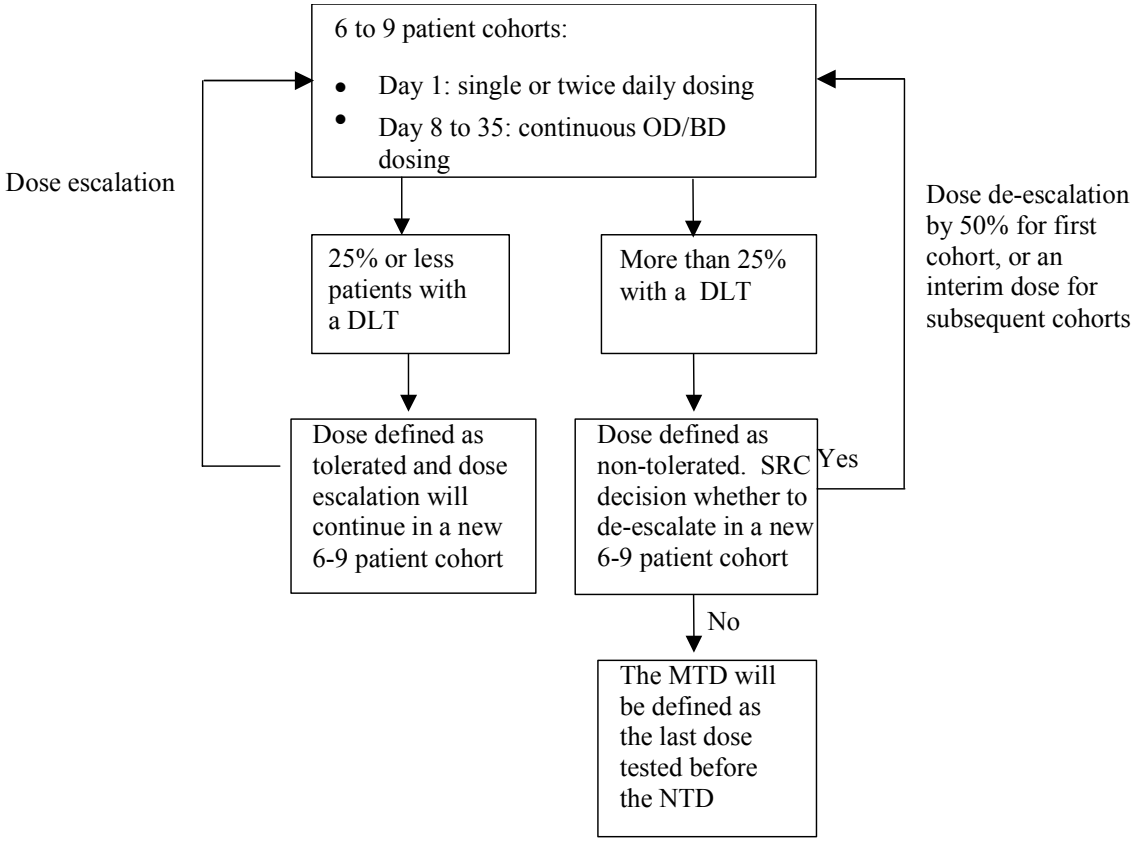

Table 1 Study plan (Part A)

Visit	**1**	**2**	**3**	**4**	**5**	**6**	**7**	**Follow up visits**			**Withdrawal from treatment[a]**	
Visit Description	**Screening visit**	**Start Tx**	**Tx**	**Tx**	**Tx**	**Tx**	**Tx**	**Tx**	**Tx**	**Tx**	**Tx stopped**	**30 days post last dose**
Day	**-14 to 0**	**1**	**8**	**15**	**22**	**29**	**36**	**57**	**NA**	**NA**		
Week	**-2 to 0**	**0**	**1**	**2**	**3**	**4**	**5**	**8**	**+ 4 [a]**	**+ 8 [a]**		
Visit Window [b] (visit date ± No. Days)	**NA**	**NA**	**± 2 days**	**± 2 days**	**± 2 days**	**± 2 days**	**± 2 days**	**± 2 days**	**± 2 days**	**± 2 days**	**NA**	**NA**
Informed consent	X											
Demography, Height, WHO performance status	X											
Medical history/Surgical history/smoking status	X											
Inclusion/exclusion criteria	X	X										
Pregnancy test (females)[d]	X	X[c]						X[d]		X[d]		
Physical examination	X	X	X	X	X	X	X	X [a]		X [a]	X	
Vital signs incl weight, and O_2 Saturation[e] measurement	X	X	X	X	X	X	X	X	X	X	X	
ECG	X[f]	X[g]			As required						X[e]	
Clinical chemistry including BNP, Haematology, Coagulation [h]	X	X[c]	X	X	X	X	X	X	X	X	X	
Urinalysis	X	X[c]	X	X	X	X	X	X	X	X	X	
RECIST evaluation	X[i]							X [j]		X [j]		
Chest X-ray [k]	X	As required										
MUGA scan/Echo[l]	X	As required					X	As required				
Ophthalmologic exam [m]	X	As required					X	As required				
PK Blood samples [n]		X		X	X	X						

Table 1 Study plan (Part A)

Visit	1	2	3	4	5	6	7	Follow up visits			Withdrawal from treatment[a]	
Visit Description	**Screening visit**	**Start Tx**	**Tx**	**Tx**	**Tx**	**Tx**	**Tx**	**Tx**	**Tx**	**Tx**	**Tx stopped**	**30 days post last dose**
Day	**-14 to 0**	**1**	**8**	**15**	**22**	**29**	**36**	**57**	**NA**	**NA**		
Week	**-2 to 0**	**0**	**1**	**2**	**3**	**4**	**5**	**8**	**+ 4** [a]	**+ 8** [a]		
Visit Window [b] (visit date ± No. Days)	**NA**	**NA**	**± 2 days**	**± 2 days**	**± 2 days**	**± 2 days**	**± 2 days**	**± 2 days**	**± 2 days**	**± 2 days**	**NA**	**NA**
PBMC blood sample [o]		X										
Optional blood sampling for host genetics research [p]		X										
Concomitant medication	X	X	X	X	X	X	X	X	X		X	X
Adverse events		X	X	X	X	X	X	X	X		X	X
Dispense study medication/check returned medication			X				X	X	X			
Diary card completion		X	X	X	X	X	X					
Part A only: Wash-out period		Days 2 to 7										

[a] Patients to attend clinic visits every 4 weeks until withdrawal from treatment (relative to Day 1). A physical examination will be performed every 8 weeks (relative to Day 1).

[b] Visit window is compared to baseline (Day 1).

[c] For Clinical chemistry, haematology, coagulation, and urinalysis repeat assessment at visit 2 is not required, if a screening assessment (visit1) was conducted within 48 hours prior to visit 2.

[d] Negative urinary or serum pregnancy test required for female pre-menopausal patients at screening, visit 2, and every 8 weeks relative to first dose with AZD8330.

[e] Any events of hypoxia demonstrated by a clinically significant drop in O2 saturations should be followed up by full PFTs and HRCT if clinically indicated.

[f] A screening ECG assessment can be conducted within 48 hours prior to visit 1.

[g] ECG measured in triplicate pre-dose and 1 hour 30 minutes and 4 hours post-dose on Day 1 and Day 15 for Part A. In addition ECGs performed in triplicate at withdrawal from treatment, any cardiac event with symptoms that may be due to cardiac ischaemia, (such as chest pain), or any arrhythmia (such as palpitations), and in cases of dyspnoea and pulmonary oedema.

[h] In the event of SAE of diarrhoea an unscheduled urea and creatinine should be performed. In the event of an adverse event associated with bleeding, a full coagulation screen including fibrinogen and d-dimer should be performed. In the event any cardiac adverse event Troponin T (or Troponin I) should be performed.

[i] Baseline tumour assessment preferably using contrast enhanced CT or MRI (see Appendix C for methods) to be performed within 4 weeks prior to first dose, (ie, repeat assessment for study is not required, if a tumour assessment was conducted as part of routine clinical practice within 4 weeks prior to starting treatment). The same method of assessment must be used at follow up.

[j] Tumour assessment preferably using contrast enhanced CT or MRI (see Appendix C for methods) should be performed every 8 weeks (relative to date of first dose with AZD8330) with a +/-7 day visit window.

[k] A chest X-Ray should be performed within 2 weeks of first dose (Day 1), and also in cases of dyspnoea or pulmonary oedema in addition to other assessments as per local standard practice.

[l] MUGA scan and/or echocardiogram performed at screening to be performed within 2 weeks prior to first dose (Day 1). Follow-up assessment is required at week 3 and week 5 (+/- 1 week). Further scans are required should signs of congestive heart failure or dyspnoea occur after this time point.

[m] A complete ophthalmologic examination must be performed within 2 weeks prior to first dose (Day 1). Follow-up assessment is required at week 5 (+/- 1 week), and ad hoc if a patient experiences a visual disturbance adverse event.

[n] Blood samples taken for PK analysis at the following timepoints: Part A: Day 1 and Day 15: Pre-dose, 15 and 30 minutes and 1, 1 hour 30 minutes, 2, 4, 6, 8, 12 (pre-dose 2nd dosing) and 24 hours post-dose (day 15 24hrs pre-dose day 16 dose). In addition: Five sparse samples on two additional days (Part A: Days 22 and 29): Pre-dose, 0.5, 1, 2 and 4 hours post-dose.

[o] Blood samples for PBMC analysis: Day 1: Pre-Dose, 30 minutes, 1 hour 30 minutes, 4, 8, and 24 hours post dose.

[p] Optional 9 mL host genetics blood samples taken prior to starting treatment (Handling instructions specified in Appendix D). If not taken at Visit 2, this sample can be taken at any visit up to withdrawal.

3.2 Rationale and risk/benefit assessment

3.2.1 Rationale for study design, doses and control groups

This study is designed to investigate the safety, tolerability and pharmacokinetic profiles of AZD8330. A dose of 0.5 mg is being proposed as the starting dose in the first-in-man study in advanced cancer patients. This is based on the FDA guidance for starting dose selection for cytotoxic agents in cancer patients, which recommends that a starting clinical dose for a 'first-in-humans' study should be either 1/10th of the severely toxic dose (STD_{10}) in rodent toxicity studies or 1/6th of the non-seriously toxic dose (NSTD) observed in non-rodent toxicity studies. Whilst a STD_{10} was not established for AZD8330 in rodents, the high dose level of 3 µmol/kg/day (8.4 mg/m^2) from the one month rat study has been used for this calculation. In dogs, the mid dose level of 0.15µmol/kg/day (1.4 mg/m^2) from the one month study represents the NSTD. Following the FDA guidance, and using the conversion factors published on the FDA website (FDA Guidance for Industry, July 2005), the human equivalent dose (HED), derived from the rat data, was calculated to be 1.4 mg/day for a 60 kg man, whilst that derived from the dog study was calculated to be 0.4 mg/day. A compromise approach to initial dose selection has been proposed and a dose of 0.5 mg/day is deemed to be an acceptable starting dose for the initial trial in patients.

Each dose escalation decision will be made after a cohort of patients have either completed a 35 day dose period on study or have experienced a DLT, at the discretion of the Safety Review Committee. Prompt pharmacokinetic analysis of samples from Day 1 will guide each dose escalation decision between cohorts, along with the review of all the safety and tolerability data. Cohorts of six patients will be recruited throughout the study to allow better characterisation of the PK and pharmacodynamic effects of AZD8330, and because when the underlying DLT rate in the population is unknown there is a lower probability of escalating from what would be a tolerated dose in the population to a non-tolerated dose using a cohort of 6 patients than with the more traditional 3 + 3 study design. To ensure there is not a consistently acute and severe tolerability issue at any new cohort, no more than 3 patients will be dosed in a cohort within the first 7 days.

Doses will be escalated by no more than 100% until the persistence of a drug related CTC Grade 2 toxicity, despite optimal therapy (apart from rash-related/dermatological toxicities, where a CTC Grade 3 will apply), after which further dose escalations will be no more than 50%. Planned rapid turnaround of pharmacokinetic samples (Day 1 and Day 15) from cohort 2 onwards will allow an assessment of potential linearity or non-linearity of the pharmacokinetics. If more information/investigation about the possible magnitude of any non-linearity observed in the pharmacokinetic data is required, the possibility exists within the protocol from cohort 2 onwards to give a lower single dose on Day 1 compared to the continuous once daily or twice daily dosing given from Day 8 onwards. In order to establish the safety and tolerability profile of AZD8330 there will be no intra-patient dose escalation of AZD8330 during the continuous once daily or twice daily dosing period in this study. Subsequent dose escalations will be within the limits outlined above, and dose escalation decisions by the Safety Review Committee will also take into account the anticipated magnitude of increase in plasma AZD8330 exposure in addition to the safety profile.

If a dose limiting toxicity (DLT) is observed in >25% patients within 35 days of commencing treatment, then this dose will be defined as a non-tolerated dose and dose escalation will stop. When the non-tolerated dose has been defined an intermediate dose(s) between the non-tolerated dose and last dose assessed may be investigated. The maximum tolerated dose will be defined as either the last dose assessed below the non-tolerated dose or a tolerated dose between these two doses.

The pharmacodynamic activity of AZD8330 will be determined by the measurement of pERK in PBMC samples from all participating patients.

The patients entering this study will have cancers refractory to standard therapies, or for whom no standard therapies exist and no control groups are planned.

No reproductive toxicology studies have been performed with AZD8330, although data from the one-month rat and dog studies indicates that AZD8330 produces adverse effects in the testes and/or ovaries in preclinical species. Therefore, AZD8330 should not be administered to pregnant or breast-feeding women and conception while on treatment must be avoided. Male patients with sexual partners who are pregnant or who could become pregnant (ie, women of child-bearing potential) should use acceptable methods of contraception (see Section 3.3.5) for 16 weeks after completing the study to avoid pregnancy and/or potential adverse effects on the developing embryo. Patients who are receiving concomitant medications that are substrates for CYP450s 3A4, 1A2 and 2C19 should be monitored for clinical signs of reduced efficacy of these agents. AZD8330 was seen to induce 3A4 and to a lesser extent CYP1A2 over the anticipated therapeutic concentration range in vitro hepatocytes studies, the results for CYP2C19 induction were inconclusive. In particular, women of childbearing potential are advised not to rely solely on hormone contraception due the potential for loss of contraceptive function as a consequence of CYP450 induction by AZD8330.

As AZD8330 is metabolised through the CYP2C9 and 2C19 pathways, it is particularly important that concomitant medications that are known inhibitors/inducers of these enzymes (http://medicine.iupui.edu/flockhart/table.htm) are recorded in order to aid the pharmacokinetic interpretation of AZD8330 data.

3.2.2 Risk/benefit and ethical assessment

There is preclinical in-vitro and in-vivo data to suggest that some tumours may be responsive to treatment with AZD8330. Preliminary Phase I data with other MEK inhibitors suggests the potential for clinical responses or prolonged stable disease in some patients for whom there are no recognised therapies (LoRusso P et al 2005, Adjei A et al 2006). The current study will recruit a similar patient population (with advanced disease, refractory to standard therapies or for which no standard therapies exist) as was recruited into the other MEK Phase I studies.

In order to preserve the safety of participating patients, we are incorporating into the study design close monitoring of all safety parameters and PK analysis in order to guide the dose escalation decisions.

The emerging pharmacokinetic (PK) and pharmacodynamic (PD) assay data, inhibition of phosphorylated ERK (pERK) in PBMCs for 0.5 to 60mg AZD8330 have been analysed in a population PK/PD model. Following administration of 20mg AZD8330 OD to steady state the population median inhibition of pERK was predicted to be approximately 80% of the baseline value at the time corresponding to the C_{max}. However, by 4h post-dose the percentage inhibition of pERK declined to approximately 40% of the baseline value. It is hypothesised that twice daily (BD) dosing will provide the potential benefit of greater duration of inhibition of the MEK pathway during the dosing interval.

Emerging safety data for AZD8330 has established an MTD of 20mg once daily (section 1.1.3). Although 40mg OD was a non-tolerated dose, the adverse events that defined its non-tolerability were generally reversible by cessation of AZD8330 and taking approximately 2 weeks to return to baseline. In general, there is precedence for drug-dose scheduling to have a significant effect on tolerability and for this reason, along with the emergent PK/PD profile, the investigation of a BD dose is considered to be clinically important and to have an acceptable risk-benefit profile.

There is currently no clinical experience with AZD8330. However, based on the preclinical safety and toxicology studies with AZD8330 and the preclinical and clinical safety profile of other MEK inhibitors, a rigorous safety evaluation package has been put in place to minimise potential risk to patients and to discover potential toxicities in a timely manner. Details of specific safety monitoring requirements (in addition to those routinely included in a Phase I oncology study) are given below and in Section 4.5.1.2.

- Soft tissue mineralisation: weekly measurement of calcium and phosphate for 5 weeks (4 weeks on part B), then 4 weekly thereafter
- APTT prolongation: weekly APTT, PT and TT measurement for 5 weeks (4 weeks in part B), then 4 weekly thereafter
- Oedema and fluid accumulation: Cardiac function assessments at baseline and during the study. Weight to be measured at every vital sign assessment. Specific analyses of urine to be done in event of peripheral oedema or persistent proteinuria.
- Dyspnoea and hypoxia: Assessments of pulse oximetry at baseline and during the study.
- Visual function disturbance: ophthalmologic examination at baseline and during the study.

Further details regarding these assessments are provided in Sections 4.5.1.2 and 4.5.2 of this protocol.

Specific inclusion / exclusion criteria and restrictions are also included in this protocol to minimise potential adverse events, for example excluding patients with a medical history of diseases associated with disorder of calcium homeostasis. Patients are also advised to refrain

from unprotected prolonged sun exposure due to the potential for phototoxicity related to AZD8330.

No reproductive toxicology studies have been performed with AZD8330, although data from the one-month rat and dog studies indicates that AZD8330 produces adverse effects in the testes and/or ovaries in preclinical species. Therefore, AZD8330 should not be administered to pregnant or breast-feeding women and conception while on treatment must be avoided. Male patients with sexual partners who are pregnant or who could become pregnant (ie, women of child-bearing potential) should use acceptable methods of contraception (See Section 3.3.5) for 16 weeks after completing the study to avoid pregnancy and/or potential adverse effects on the developing embryo. Women of childbearing potential are advised not to rely solely on hormone contraception due the potential for loss of contraceptive function as a consequence of CYP450 induction by AZD8330.

Given the extensive safety monitoring included in this study, and inclusion of a full review of safety, tolerability and pharmacokinetic data prior to each dose escalation decision, AstraZeneca considers the overall risk for the patients who participate in this study to assess the safety and tolerability of AZD8330 is acceptable due to the lack of highly effective alternative treatments in this patient population.

3.3 Selection of study population

3.3.1 Study selection record

Investigator(s) must keep a record of patients who were considered for enrolment but were never enrolled eg, 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 patients must fulfil all of the following criteria:

1. Provision of informed consent.

2. Male or female, aged 18 years or older.

3. Cancer which is refractory to standard therapies, or for which no standard therapies exist.

4. Measurable Lesions that can be accurately measured in at least one dimension (longest diameter to be recorded) as ≥20mm with conventional techniques or as ≥10mm with spiral CT scan or non measurable lesions according to the RECIST Criteria.

5. Laboratory value as listed below: (SI Units).

- Calculated serum creatinine clearance ≥50 mL/min (using Cockcroft-Gault formula (see Appendix E) or by 24 hour urine collection).

6. LVEF as determined at baseline MUGA/echocardiography at or above the normal level.

7. WHO performance status 0-2 (those with performance status 2 must have been stable with no deterioration over the previous 2 weeks).

8. Evidence of post-menopausal* status or negative urine or serum pregnancy test for female pre menopausal patients.

* Post menopausal females are defined as follows: Natural menopause with menses >1 year ago; radiation induced oophorectomy with last menses >1 year ago; chemotherapy induced menopause with 1 year interval since last menses; serum FSH and LH and plasma oestradiol levels in the postmenopausal range for the institution; bilateral oophorectomy or hysterectomy.

3.3.3 Inclusion criteria for optional host genetic research

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

1. Provision of informed consent for host genetic sampling (blood sample) and analyses.

If a patient declines to participate in the genetic research (see Appendix D), 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, so long as they consent to the main study.

3.3.4 Exclusion criteria

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

1. Laboratory values as listed below: (SI Units)

   - Absolute Neutrophil Count (ANC) <1.5 x 10^9/L (1500 per mm^3)
   - Platelets <100 x 10^9/L (100,000 per mm^3)
   - Haemoglobin (Hgb) <9.0 g/dL
   - Serum bilirubin ≥1.5x Upper Limit of Normal (ULN)
   - Aspartate aminotransferase (AST/SGOT) or alanine aminotransferase (ALT/SGPT) ≥2.5 x ULN

- Serum calcium (corrected for albumin[1]) outside of normal limits
- Phosphate outside of normal limits
- Serum potassium or magnesium outside of normal limits

2. Any radiotherapy, biological or chemotherapy within 21 days prior to starting the study (not including palliative radiotherapy at focal sites) or who have not recovered from side effects of their treatment

3. Participation in an investigational drug study within the 30 days prior to entry or who have not recovered from clinically significant side effects of an investigational study drug

4. Brain metastases or spinal cord compression unless treated and stable off steroids and anticonvulsants (for at least 1 month)

5. Female patients who are breast feeding, or patients of reproductive potential not employing an effective method of birth control

6. Any evidence of severe or uncontrolled systemic disease (e.g., severe hepatic impairment, severe renal impairment uncontrolled diabetes, acute uncontrolled infection) or current unstable or uncompensated respiratory or cardiac conditions or peripheral vascular disease including diabetic vasculopathy

7. Parathyroid disorder or history of malignancy associated hypercalcaemia

8. Evidence of active infection or active bleeding diatheses

9. Recent major surgery within 4 weeks prior to entry into the study (excluding the placement of vascular access) which would prevent administration of study treatment

10. Patients with documented cases of human immunodeficiency virus (HIV) or hepatitis B or C

11. Refractory nausea and vomiting, chronic gastrointestinal diseases (eg, inflammatory bowel disease), or significant bowel resection that would preclude adequate absorption

12. Previous treatment with a MEK inhibitor

13. QTc interval > 450ms

[1] Corrected calcium (mmol/l) = Total calcium (mmol/l) + 0.02 x [40 – Serum albumin (g/l)]

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

15. Previous enrolment or randomisation of treatment in the present study

16. Clinical judgement by the Investigator that the patient should not participate in the study

3.3.5 Restrictions

- Females of childbearing age are required to use reliable contraception throughout dosing and for 4 weeks post dosing.

- All males with partners of childbearing potential or whose partners are pregnant must use barrier contraception for the duration of dosing and for 16 weeks post dosing.

- Reliable methods of contraception should be used consistently and correctly and acceptable methods include barrier methods, implants, injectables, some IUDs, sexual abstinence or vasectomised partner. It is possible that AZD8330 interacts with oral contraception so any female using oral contraception must also use a barrier method of contraception. Hormonal contraception alone is not acceptable.

- No Grapefruit or grapefruit juice to be consumed.

- AZD8330 should be taken on an empty stomach (no food or drink other than water for 2 hours prior to dosing and 1 hour after dosing).

- Avoid excessive sun exposure and use adequate sunscreen protection if sun exposure is anticipated.

- Patients who are receiving concomitant medications that are substrates for CYP450s 3A4, 1A2 and 2C9 should be monitored for clinical signs of reduced efficacy of these agents (http://medicine.iupui.edu/flockhart/table.htm). In particular, patients who are on anticoagulant therapy (eg, warfarin) should have their anticoagulation tested more frequently when receiving AZD8330.

3.3.6 Discontinuation of patients from treatment or assessment

3.3.6.1 Criteria for discontinuation

Patients may be discontinued from study treatment and assessments at any time. Specific reasons for discontinuing a patient from this study are:

- 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 ie, the patient does not meet the required inclusion/exclusion criteria for the study (see Section 3.3.6.3)
- Patient lost to follow-up
- A female patient becoming pregnant
- Disease progression
- Deterioration in the patients condition which in the opinion of the Investigator warrants study medication discontinuation (to be recorded as an AE or under Investigator Discretion)
- Adverse Event

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

- Withdrawal of consent for host genetics research. A patient may withdraw from host or tumour 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

3.3.6.2 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 assessed by an investigator(s). Adverse events should be followed up.

Any serious adverse event (SAE) or non-serious AE considered related to study treatment by the investigator, which is ongoing at patient discontinuation from the study, or occurs within 30 days of the last study treatment must be followed up to resolution, unless the SAE is considered by the investigator to be unlikely to resolve.

AstraZeneca reserves the right to ask for further information/clarification on any AE that may be considered of interest.

3.3.6.3 Procedures for discontinuation from optional tumour biomarker aspects of the study (Part B only) (Not applicable)

No longer applicable.

3.3.6.4 Procedures for handling incorrect enrolled patients

Patients not meeting the inclusion/exclusion criteria for the study should, under no circumstances, be enrolled into the study - there can be no exceptions to this rule.

Where patients not meeting the study criteria are enrolled in error, or where patients subsequently fail to meet the criteria for the study post enrolment, the investigator will in conjunction with AstraZeneca discuss whether such a patient should withdraw from study taking into consideration ethical and safety factors and the discussion and rationale for each decision will be documented.

Incorrectly enrolled subjects will be listed, and only included in analysis if subjects' data were used for dose escalation decision.

3.3.6.5 Procedures for discontinuation from host 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 host genetic research. It must be established whether the patient:

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

- Withdraws consent for the sample to be kept for host 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 host 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 host 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

AstraZeneca Investigational Products section will supply AZD8330 open labelled clinical study material.

Table 2 Identity of investigational product

Investigational product	Formulation	Dosage form and strength	Manufacturer	Formulation number [a]
AZD8330	Film Coated Tablets	0.5 mg	AstraZeneca	F013509
AZD8330	Film Coated Tablets	0.75 mg	AstraZeneca	F013526
AZD8330	Film Coated Tablets	1.5 mg	AstraZeneca	F013511
AZD8330	Film Coated Tablets	5.0 mg	AstraZeneca	F013513

[a] Batch numbers for all investigational products will be identified in the clinical study report

3.4.2 Doses and treatment regimens

3.4.2.1 Tablets

AZD8330 tablet formulation drug product will be supplied in 0.5 mg, 0.75 mg, 1.5 mg and 5 mg strengths in high-density polyethylene (HDPE) bottles, each bottle will contain sufficient tablets for each patient treatment period, as appropriate. The Safety Review Committee will give dose recommendations, and patients will be required to take the same combination of tablet strengths to make up the relevant dose, as directed by AstraZeneca.

If patients are not able to tolerate AZD8330, dose reductions are permitted as described in Section 3.1.4.1. This will depend on doses available and doses selected for the oral formulation.

3.4.2.2 Part A (Dose Escalation Phase)

The first cohort will receive a single 0.5 mg dose of AZD8330 on Day 1, followed by 0.5 mg once daily (OD) continuous dosing from Day 8 onwards. Subsequent cohorts will be dispensed investigational product according to the dose directed by the Safety Review Committee (see Section 3.1.4.1). Each dose should be taken with approximately 240 ml (8 fl oz) water, and on an empty stomach no food or drink other than water for 2 hours prior to dosing and 1 hour after dosing (see Section 3.3.5).

3.4.2.3 Part B (Safety Expansion Phase) (Not applicable)

No longer applicable.

3.4.3 Labelling

Each bottle of AZD8330 Tablets 0.5 mg, 0.75 mg, 1.5 mg and 5 mg will be labelled by Investigational Products (IPS), AstraZeneca, or CRO.

Each bottle of AZD8330 will have a label permanently affixed to the outside and will be labelled in accordance with Good Manufacturing Practice and local regulations, stating that the material is for clinical trial / investigational use only and should be kept out of reach of

children. Labels will include blank lines for the patient enrolment code (E-code) and date dispensed.

3.4.4 Storage

All investigational products must be kept in a secure place under appropriate storage conditions. Descriptions of the appropriate storage conditions are specified on the investigational products label. The tablets should all be stored in their original packaging until use.

3.4.5 Accountability

It is the Investigator/institution's responsibility to establish a system for handling study treatments, including investigational products, so as to ensure that:

- deliveries of such products from AstraZeneca are correctly received by a responsible person (eg, a pharmacist)
- deliveries are recorded
- study treatments are handled and stored safely and properly
- study treatments are dispensed only to study patients in accordance with the protocol
- patients return all unused medication and empty containers to the Investigator.

At the end of the study, it must be possible to reconcile delivery records with records of usage and destroyed/returned stock. Records of usage should include the identification of the person to whom the study treatment was dispensed, the quantity and date of dispensing, and unused study treatment returned to the Investigator. Certificates of delivery and return must be signed, preferably by the Investigator or a pharmacist, and copies retained in the Investigator File.

All unused or returned medication, after drug accountability, should be destroyed at site wherever possible. In the event that this is not possible the medication should be returned to the local distribution centre for destruction.

3.5 Method of assigning patients to treatment groups

Written informed consent will be obtained before enrolment, when patients will be identified with a unique 7-digit enrolment number (E00NNXXX: NN being the centre number, XXX being the patient enrolment number at the centre.). Enrolment numbers will start at 999 in each centre and go down (eg, at centre 1, patients will be assigned e-codes E0001999, E0001998, E0001997, etc and E0002999, E0002998, E0002997, etc in centre 02).

Part A (Dose Escalation Phase)

Patient eligibility will be established before assigning a study patient number starting with number 101 in Part A. Patient numbers will be assigned strictly sequentially, by a central co-ordinator at AstraZeneca as patients are confirmed as eligible to receive treatment. If a patient discontinues from the study, the patient number will not be reused, and the patient will not be allowed to re-enter the study.

Replacement patients may enrol until the minimum required number of evaluable patients is available for assessment. Reasons for patient withdrawal will be given due consideration when enrolling patients.

3.6 Pre-study, concomitant and post-study treatment(s)

The following treatment/drugs are restricted in this study;

- No other chemotherapeutic agents, or investigational drugs should be administered whilst patients are receiving study medication.
- Any radiotherapy, biological or chemotherapy within 21 days prior to starting the study (not including palliative radiotherapy at focal sites).
- Recent major surgery within 4 weeks prior to entry into the study (excluding the placement of vascular access) which would prevent administration of study treatment.
- Patients who are receiving concomitant medications that are substrates for CYP450s 3A4, 1A2 and 2C9 should monitor for clinical signs of reduced efficacy of these agents (http://medicine.iupui.edu/flockhart/table.htm). In particular, patients who are on anticoagulant therapy (eg, warfarin) should have their anticoagulation tested more frequently when receiving AZD8330.
- Concomitant medications that are known inhibitors/inducers of CYP2C9 and 2C219 pathways (http://medicine.iupui.edu/flockhart/table.htm) should be recorded (dose, frequency and dates of dosing) in order to aid the pharmacokinetic interpretation of AZD8330 data.

Other medication that is considered necessary for the subject's safety and well-being may be given at the discretion of the investigator(s). The administration of all medication (including investigational products) must be recorded in the appropriate sections of the pCRF.

It is advisable that during the PK sampling periods of Part A, patients should avoid changes to or the addition of all concomitant medications, in particular any that are likely to affect the metabolism of AZD6244 (eg, CYP1A2 or 3A4 inducers), unless considered clinically essential for management of concurrent conditions.

3.7 Treatment compliance

Administration of study drug will be captured in the pCRF.

Study treatments are only dispensed to patients in accordance with the protocol. Patients must return all unused medication and packaging. At the end of the study, it must be possible to reconcile delivery records with records of usage and returned stocks. Any discrepancies must be accounted for. Certificates of delivery and return must be signed, preferably by the Investigator or a pharmacist.

4. MEASUREMENTS OF STUDY VARIABLES AND DEFINITIONS OF OUTCOME VARIABLES

4.1 Primary variable

The primary variables are incidence and severity of adverse events, clinical chemistry, haematology and urinalysis, vital signs, MUGA/Echo parameters, ECG parameters, ophthalmologic examinations and O_2 saturation.

4.2 Screening and demographic measurements

Prior to enrolment in the study, each patient will undergo an enrolment examination within 2 weeks prior to visit 2 (Day 1). This will consist of:

- Provision of written informed consent
- Evaluation against inclusion and exclusion criteria
- Recording of demographic data – date of birth, sex, height, weight, race
- Standard medical and surgical history and a physical examination.
- Smoking status
- Resting supine blood pressure (BP) and pulse measurement
- Pulse oximetry
- Full ophthalmological exam
- WHO performance status
- ECG
- Blood sample for standard clinical chemistry (including BNP), coagulation, and haematology assessments

- Urine sample for urinalysis
- MUGA Scan or echocardiogram
- Chest x-ray
- Pregnancy test (urine or serum) for female pre menopausal patients
- Baseline tumour assessment according to the Response Evaluation Criteria In Tumours' (RECIST) criteria (see Appendix C) in RECIST evaluable tumours within 4 weeks of dosing.
- Recording of any concomitant medications.

4.3 Pharmacokinetic measurements and variables

The methods for collection of biological samples and derivation of PK parameters are presented below and in the Laboratory Handbook for Investigators.

Depending on emerging data, the actual timings of these samples may change, but the total blood volume will not change. In order to fully elucidate the terminal elimination phase following a single dose of AZD8330 on Day 1 in Part A, if warranted by the data PK sampling may be redistributed be cover a 48 or 72h period (ie total number of samples will not change). If this occurs, dosing on these subsequent days (ie Days 2 and possibly 3) should be withheld.

Part A: Pharmacokinetic sampling

Blood samples for the determination of plasma AZD8330 will be taken on Days 1 and 15 as described below:

- Day 1: Pre-dose, 15 and 30 minutes and 1, 1 hour 30 minutes, 2, 4, 6, 8,12 (pre-dose 2nd dosing) and 24 hours post dose.
- Day 15: Pre-dose, 15 and 30 minutes and 1, 1 hour 30 minutes, 2, 4, 6, 8,12 (pre-dose 2nd dosing) and 24 hours post-dose (pre-dose Day 16 dose).
- Five sparse sampling on two additional days at steady-state, Days 22 and 29: pre-dose, 0.5, 1, 2 and 4 hours post-dose.

In addition to AZD8330, other major metabolites may be analysed from the same blood and urine samples if warranted and if a bioanalytical assay is available.

4.3.1 Collection of biological samples

Analysis of plasma samples for the determination of AZD8330 and any known major metabolite will be the responsibility of Drug Metabolism and Pharmacokinetic Department, Alderley Park, AstraZeneca, UK.

Venous blood samples (2 mL) will be collected to provide plasma for analysis.

Labels for tubes are unique, enabling identification of the study, compound to be analysed, patient/centre number, visit number and protocol scheduled time point.

Samples will be shipped initially to the coordinating laboratory, and subsequently sent for analysis to a referral laboratory. A sample inventory must accompany each shipment of samples.

Details of sample collection, storage and shipment will be provided in the Laboratory Manual for Investigators.

Although every attempt should be made to collect all samples as protocolled, it is accepted that this will not always be possible and therefore it is essential that the actual time and date of collection of each blood sample (whether collected as protocolled or not) is recorded on the pCRF.

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

Blood samples (2 mL) for determination of plasma AZD8330 will be taken at the times specified in the Study Plan Table 1 and Section 4.3. The actual timings may change depending on the emerging data from this study. Other metabolites may be analysed from the same blood sample if warranted. PK samples will not exceed the volumes provided in Table 3. The date and time of collection will be recorded. Further details of sample collection and shipment will be provided in the Laboratory Manual for Investigators.

The pharmacokinetic analyses will be undertaken for both plasma AZD8330 and any known major metabolite by (or on behalf of) the Clinical Pharmacology Group, AstraZeneca (Alderley Park).

4.3.2.1 Non Compartmental Analysis

The maximum plasma concentrations (C_{max}) and the time to reach the maximum plasma concentrations (t_{max}) will be determined by visual inspection of the plasma concentration-time profiles. Where more than one maximum occurs, the reported value will be assigned to the first occurrence. The area under the plasma concentration-time curve from zero to the time of the last quantifiable plasma concentration, AUC(0-t), will be calculated by the linear trapezoidal rule. For the first single dose, the area from the last quantifiable drug concentration to infinite time, AUC, will be calculated by the following formula:

AUC(0-t)+ $\frac{Ct}{\lambda z}$, where Ct is the last quantifiable concentration and λ_z is the terminal rate constant, calculated by log-linear regression of the terminal portion of the concentration-time profile where there are sufficient data (ie, there are at least 3 points in the terminal phase). The terminal half-life ($t_{½}$) will be calculated from the equation $\ln(2)/\lambda_z$. If the data allow, the

apparent oral clearance (CL/F), the apparent volume of distribution based on the terminal phase (Vz/F) and at steady-state Vss may be calculated. Following multiple doses, the AUC over the dosing period (AUC_{τ}), will be calculated. If the data allow, the accumulation index may also be calculated.

4.3.2.2 Compartmental Analysis (Not applicable)

Not applicable.

4.3.2.3 Pharmacokinetic/Pharmacodynamic modelling

If the data are suitable the relationship between the plasma AZD8330 and any known major metabolite concentrations/exposure and changes in pharmacodynamic and safety profile will be investigated either using graphically means or appropriate PK/pharmacodynamic software.

4.4 Efficacy and pharmacodynamic measurement and variables

4.4.1 Tumour assessment by imaging techniques using RECIST

The RECIST guidelines for measurable, non-measurable, target and non-target lesions, and the objective tumour response criteria (complete response (CR), partial response (PR), Stable disease (SD) or progression of disease (PD)) are presented in Appendix C. The RECIST criteria will be used to programmatically determine ORR. Patients with non-measurable disease only will be assessed according to RECIST criteria for non-target and new lesions in Appendix C.

Baseline tumour assessments should be performed no more than 4 weeks before the start of study treatment, but should be as close as possible to the start of study treatment. Following date of the first dose with AZD8330, efficacy for all patients will be assessed by objective tumour response every 8 weeks thereafter relative to the date of the first dose with AZD8330. Assessments may be performed +/- 7 days relative to the specified visit date. In addition, patients who progress between scheduled visits should have a radiological imaging for confirmation.

Baseline radiological examination, preferably contrast enhanced CT, will be performed on anatomical coverage to adequately define all areas of disease. Post-baseline imaging should follow and evaluate all lesions identified at baseline. MRI should only be used where CT is not feasible or it is medically contra-indicated.

For patients with measurable lesions, all measurable lesions confirmed and assessed by radiological methods (CT or MRI scans) up to a maximum of 5 lesions per organ and 10 lesions in total, representative of all involved organs, should be identified as target lesions, recorded and measured at baseline, and at the time points specified above.

Non-target lesions will also be monitored throughout the study, and an assessment of non-target lesions will be made and recorded as “present”, “present with progression” or “absent”. Details of any new lesions will also be collected.

If an unscheduled radiological and clinical tumour assessment is performed, and the patient has not progressed, the next scheduled tumour assessment should still be performed at the planned time (as detailed in the study plan) relative to date of first dose of AZD8330.

Tumour assessment will be performed in accordance with the protocol schedule until evidence of one of the following:

- Progression of disease (patients will be followed for subsequent therapy and survival)
- Death without evidence of progression
- Withdrawal of consent
- Withdrawal from treatment

If a patient has any palliative radiotherapy to a lesion that lesion should not be included in assessment of response, but should be assessed for progression.

A patient will be determined to have progressed if they have progression of target lesions, clear progression of existing non-target lesions, or the appearance of one or more new lesions. Death will be regarded as a progression event in those patients who die before disease progression. Unequivocal malignant disease not identified prior to starting study treatment on additional anatomical imaging (e.g., computed tomography (CT), magnetic resonance imaging (MRI) or bone scan confirmed by X-ray), prompted by symptoms is considered disease progression and should be recorded as new lesions. If progression is uncertain, patients may continue on treatment until the next scheduled assessment (ie, 8 weeks later +/- 1 week) or may have an unscheduled assessment earlier than this if considered appropriate by the investigator.

Lesions must be assessed using the same method and technique on each occasion. Lesions will be recorded on the pCRF page in the same order as they were recorded at screening. Details of any new lesions will also be collected. Response will be calculated in comparison to the baseline tumour measurements obtained before starting treatment. Progression will be calculated in comparison to when the tumour burden was at a minimum. Overall visit response will be recorded on the pCRF.

4.4.2 Objective response rate (including RECIST)

4.4.2.1 Methods of assessment

Patients with measurable disease at baseline will be categorised using RECIST.

Patients with non-measurable disease at baseline will be assessed for CR, SD and PD.

Categorisation of overall visit response will be based on RECIST using the following response categories: CR, PR, SD, and PD (see Appendix C). In the case of stable disease,

measurements must have met the stable disease criteria at least once after randomization for a minimum interval of 6 weeks.

To be assigned a status of PR or CR, changes in tumour assessments must be confirmed no less than 4 weeks after the criteria for response were met.

4.4.2.2 Derivation or calculation of outcome variable

Best overall response will be calculated as the best response recorded from date of randomisation (taking as reference for progressive disease the smallest measurements recorded since the treatment started) for each patient, and will be used for the summaries of objective response. Best overall response will be determined programmatically based on the RECIST criteria.

ORR is defined as the proportion of patients who have a best response of either CR or PR.

4.4.3 Tumour assessments for patients with non-measurable disease at baseline

Patients with non-measurable disease are not excluded from this study. These patients should be followed up with the same assessment schedule as those with measurable disease at baseline and outlined in Section 4.4.1 preferably using contrast enhanced CT or MRI where this is not feasible (see Appendix C for methods).

4.4.3.1 Methods of assessment

Assessments of these patients will be under investigators discretion in line with their institutional policies.

4.4.4 Phospho-ERK in PBMCs

4.4.4.1 Methods of assessment

The level of ERK phosphorylation (pERK) will be assessed in PBMCs by measuring phorbol ester (TPA) stimulated pERK production, using specific antibodies and flow cytometry.

All patients will contribute blood samples for PBMC preparation at each of the following time points:

- Day 1: Pre-dose, 30 minutes, 1 hour 30 minutes, 4, 8 and 24 hours

At each time point 2 x 2mL of blood will be drawn for PBMC analysis. Drawn samples will be immediately processed, stored and then shipped as detailed in the Laboratory Handbook for Investigators. Samples will be shipped initially to the coordinating laboratory, and subsequently sent for analysis to a referral laboratory.

4.4.5 Phospho-ERK in matched tumour biopsies (Not applicable)

No longer applicable.

4.4.5.1 Methods of assessment (Not applicable)

No longer applicable.

4.4.6 Other biomarker analysis of tumour biopsy material (Not applicable)

No longer applicable.

4.4.6.1 Methods of Assessment (Not applicable)

No longer applicable.

4.5 Safety measurements and variables

The methods for collecting safety data are described below.

4.5.1 Adverse events

4.5.1.1 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.

4.5.1.2 Recording of adverse events

AEs/SAEs will be collected throughout the study; from informed consent until 30 days after study treatment is discontinued. SAEs occurring in the 30-day follow up period should be reported to AstraZeneca in the usual expedited manner.

If a patient withdraws from treatment for reasons other than disease progression and therefore continues to have tumour assessments using RECIST, drug or procedure related SAEs must be captured until the patient is considered to have progressive disease, and therefore will have no further RECIST assessments.

During the course of the study all AEs and SAEs should be proactively followed up for each patient; events should be followed up to resolution, unless the event is considered by the investigator to be unlikely to resolve due to the underlying disease. Every effort should be made to obtain a resolution for all events, even if the events continue after discontinuation/study completion.

Any SAE or non-serious AE considered related to study treatment by the investigator, that is ongoing when the patient completes the study or at patient discontinuation from the study, or occurs within 30 days following treatment discontinuation, must be followed up to resolution, unless the AE is considered by the investigator to be unlikely to resolve, or the subject lost to follow up.

AstraZeneca reserves the right to ask for further information/clarification on any AE that may be considered of interest.

At each visit the method of detecting AEs and SAEs in this study will be by:

(a) information volunteered by the patient, the patients parent or carer

(b) open-ended non-leading verbal questioning of the subject such as the following: "Have you had any health problems since the previous visit?"

(c) observation by the investigational team, other care providers or relatives

A description of the event will be provided, the dates of onset and resolution. If a diagnosis of the patient's condition has been made, then the diagnosis should be recorded as the AE. However if the diagnosis of the patient's condition has not been made, or only if the individual symptoms are not well recognised, then the individual symptoms should be recorded separately.

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 4.5.1.1. 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 graded according to the National Cancer Institute Common Terminology Criteria for AEs (NCI CTC Version 3, June 2003).

The following variables will be collected for each AE:

- AE description
- Onset date and time
- Resolution date and time
- Changes in CTC grade
- Action taken
- Treatments patient received for AE
- Outcome
- Causality (yes or no)

- Whether event constitutes an SAE

AEs will be coded using MEDRA (Medical dictionary for regulatory activities).

After study completion (ie, after any scheduled follow-up period has ended) there is no obligation to actively report information on new AEs or SAEs occurring in former study patients. However if an investigator learns of any SAEs, including death, at any time after a patient has completed the study and he/she considers there is a reasonable possibility that the event is related to AZD8330, the investigator should notify AstraZeneca, Drug Safety.

Abnormalities in laboratory data

The reporting of protocol mandated laboratory/vital sign abnormalities as both laboratory findings and AEs should be avoided. They should not be reported as AEs unless any criterion for an SAE is fulfilled, the laboratory/vital sign abnormality causes the patient to discontinue treatment with the investigational product (IP) or if the investigator has a strong belief that it should be reported as an AE. All abnormalities from protocol-mandated laboratory/vital measurements will summarised in the Clinical Study Report in the 'Laboratory measurements and variables' section.

If an abnormal laboratory value/vital sign is associated with a diagnosis or clinical signs or symptoms, then the diagnosis, sign or symptom should be reported as an AE and the associated laboratory result/vital sign should be considered additional information to support the diagnosis. This applies to both protocol-mandated measurements and those that are measured outside of protocol requirements.

Disease progression

Disease progression can be considered as a worsening of a patient's condition attributable to the disease for which the IP is being studied. It may be an increase in the severity of the disease under study (DUS) and/or increases in the symptoms of the disease. Expected progression of the DUS and/or expected progression of signs and symptoms of the DUS, unless more severe in intensity or more frequent than expected for the patient's condition, should not be reported as an AE. The development of new, or progression of existing metastasis to the primary cancer under study should be considered as disease progression and not an AE. Events which are unequivocally due to disease progression, must not be reported as an AE/SAE.

Lack of efficacy

Where there is deterioration in the condition for which the study treatment is being used, there may be uncertainty as to whether this is lack of efficacy, disease progression or constitutes an AE. In such cases, unless the AstraZeneca or reporting physician considers that the study treatment contributed to the deterioration, or local regulations state to the contrary, the deterioration should be considered to be lack of efficacy and not an AE.

Handling of deaths

All deaths that occur during the study, or within the 30-day follow-up period after the administration of the last dose of study treatment, must be reported as follows:

- Death which is clearly as a result of disease progression should be communicated to the study monitor at the next monitoring visit and should be documented in the pCRF module, but should not be reported as a SAE

- Where death is not due (or not clearly due) to progression of disease under study the AE causing the death must be reported to the study monitor as an SAE within 24 hours. The report should contain a comment regarding the co-involvement of progression of disease, if appropriate, and should assign a single primary cause of death together with any contributory causes

- Deaths with an unknown cause should always be reported as a SAE but every effort should be made to establish a cause of death. A post-mortem may be helpful in the assessment of the cause of death, and if performed a copy of the post-mortem results (with translation of important parts into English) should be forwarded to AstraZeneca, Drug Safety within the usual timeframes

New cancers

The development of a new cancer should be regarded as an AE and will generally meet at least one of the serious criteria. New cancers are those that are not the primary reason for the administration of the study treatment and have been identified after the patient's inclusion in this study. They do not include metastases of the original cancer. Symptoms of metastasis or the metastasis itself should not be reported as an AE/SAE as they are considered to be disease progression.

Cardiac Dysfunction

For patients who develop symptoms consistent with cardiac dysfunction (e.g. congestive cardiac failure, dyspnoea or peripheral oedema) a measure of cardiac ejection fraction (LVEF) and blood sample for Troponin T (or Troponin I) (clinical chemistry sample) and BNP or NT-pro BNP should be taken at baseline and at the time of the event. All events of dyspnoea or pulmonary oedema should be followed up with an ECG and chest X-ray, and any events of hypoxia demonstrated by a clinically significant drop in O_2 saturations should be followed up by full PFTs and HRCT if clinically indicated.

Visual Disturbances

All events of visual disturbance should be followed up with a complete ophthalmological examination, including slit-lamp examination.

Diarrhoea

All events of diarrhoea should be followed up with a complete electrolyte assessment.

Fluid accumulation

All events of peripheral oedema should be followed up with clinical chemistries (including BNP, electrolytes and albumin) and routine urinalysis. For patients who develop other fluid accumulation conditions such as ascites or pleural effusion (or experience worsening of pre-existing condition), follow-up should include measurement of clinical chemistries (including electrolytes and albumin) and urinalysis.

Adverse events associated with bleeding

For patients who develop adverse events associated with bleeding, such as haematochezia or haematemesis, a full coagulation screen should be performed including APTT, PT, TT, fibrinogen and d-dimer.

Proteinuria

For patients who develop proteinuria (or experience worsening of pre-existing condition), a repeat urinalysis should be performed. If this second test confirms the laboratory abnormality the full 24-hour urine collection should be performed to measure urinary protein excretion.

Overdose

To date, no subject has experienced an overdose with AZD8330. There is currently no known antidote to AZD8330. The treatment of AEs associated with overdose should be supportive for the underlying symptoms. Doses of study treatment in excess of that specified in the clinical study protocol are considered to be an overdose.

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.

Pregnancy

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.

4.5.1.3 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 (ie, 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.

4.5.2 Laboratory safety measurements and variables

4.5.2.1 Methods of assessment

The following clinical chemistry, urinalysis and haematology tests will be performed:

Clinical chemistry	**Haematology**
s - Albumin	B - Erythrocyte count
s - Alanine aminotransferase (ALT)	B - Haemoglobin
s - Aspartate aminotransferase (AST)	B - Platelet count
s - Alkaline phosphatase (ALP)	B - Leucocyte cell count
s - Total Calcium	B - Leucocyte differential count (absolute count):
s - Creatinine	B - Monocytes,
s - Gamma glutamyltranserase (γGT)	B - Neutrophils,
s - Glucose	B - Lymphocytes,
s - Magnesium	B - Basophils
s - Phosphate	B - Eosinophils
s - Potassium	B/p - Activated Partial Thromboplastin Time (APTT)
s - sodium	B/p-Prothrombin Time (PT)
s - Total protein	B-Thrombin Time (TT)
s - Total bilirubin	
s - Urea nitrogen	
s/p - BNP or NT-proBNP	
s-Troponin T (or Troponin I)	
Urinalysis	
U-Glucose	U-Blood
U-Protein	U-Microscopy (red blood cells and white blood cells, bacteria, casts and crystals) only perform if urinalysis abnormal

An unscheduled serum urea and creatinine test should be performed in every case of an SAE of diarrhoea. In addition, clinical chemistry, haematology, coagulation and urinalysis testing will be repeated as clinically indicated as part of the routine management of the patient on the occurrence of AEs.

Adjusted Calcium will be calculated using for formula below, from Total Calcium and Albumin:

Adj. Calcium = Total Calcium (mmol/L) + ([40 – Albumin (G/L)] x 0.02)

4.5.3 Vital signs, ECG and physical examination

4.5.3.1 Vital signs

Resting supine blood pressure and pulse rate will be measured using a semi-automatic blood pressure recording device with an appropriate cuff size. See study plan Table 1 for time points.

Any symptoms from the patient should if applicable be registered as an AE.

4.5.3.2 ECG

(a) Screening (all patients)

All patients participating in the study will have a screening ECG. Patients will be excluded from the study if a QTc is >450 ms. The screening ECG will be analysed locally. The screening assessment can be conducted within 48 hours of visit 1.

(b) Treatment Phase ECGs

Patients receiving AZD8330 will have 12-lead ECGs captured in triplicate pre-dose, 1 hour 30 minutes and 4 hours post-dose on Visit 2 (Day 1), and Day 15. In addition ECGs performed in triplicate at withdrawal from treatment, any cardiac event with symptoms due to cardiac ischemia (such as chest pain, or any arrhythmia (such as palpitations), and in cases of dyspnoea and pulmonary oedema.

All ECGs (with the exception of screening ECGs) will be transmitted electronically for central reading and interpretation by an AstraZeneca appointed external cardiologist. Details of recording and transmission of ECGs are provided in the study specific ECG manual provided to investigators. Parameters including QRS duration, R-R, PQ, and QTc will be determined and reviewed by an external cardiologist. Pulse rate and QTcF, will be calculated by AstraZeneca from the data provided by the external cardiologist.

Patients who continue to receive treatment beyond the defined end of study, in accordance with Section 7.6 (Study timetable and end of study) will no longer have ECGs analysed by an external cardiologist, but should continue to be reviewed by site staff as per routine practice.

The Investigator should review the paper copy of the ECGs on each study day and may refer to a local cardiologist if appropriate.

Any symptoms from the patient should be registered as a comment and if AE criteria are met, recorded as an AE.

4.5.3.3 Physical examination

A physical examination will be performed at screening, weekly up to and including week 4, week 5, week 8, and every 8 weeks thereafter. Details will be recorded in the pCRF.

4.5.4 Other safety measurements and variables

4.5.4.1 Pregnancy test

A pregnancy test (urine or serum) will be performed at screening, prior to starting treatment, and every 8 weeks relative to first dose with AZD8330 for female pre-menopausal patients.

4.5.4.2 MUGA scan/Echocardiogram

A MUGA and/or Echocardiogram will be conducted at screening, week 3 and week 5 (+/- 1 week). In addition, MUGA or Echocardiogram scans are required should signs of congestive heart failure occur at any time.

The modality of the cardiac function assessments must be consistent within patient, ie, if echocardiogram is used for the screening assessment then echocardiogram should also be used for subsequent scans if required.

4.5.4.3 BNP or NT-proBNP

Either BNP or NT-proBNP will be tested locally according to local practice at baseline (Visit 2) prior to initiation of treatment and thereafter at the same time as safety labs (see Table 1). In addition an unscheduled sample should be taken if there are any signs of congestive heart failure or dyspnoea.

The modality of these blood assessments must be consistent within patient, ie, if BNP is used for the screening assessment then BNP should also be used for subsequent assessments.

4.5.4.4 Troponin T (or Troponin I)

Troponin T (or Troponin I) will be tested at baseline (Visit 2) prior to initiation of treatment and thereafter at the same time as other safety labs (see Table 1) as part of the clinical chemistry sample. In addition an unscheduled sample for Troponin T (or Troponin I) analysis should be taken if there are any signs of cardiac adverse events.

4.5.4.5 Coagulation screen

APTT, PT and TT will be tested at baseline (Visit 2) prior to initiation of treatment and thereafter at the same time as other safety labs (see Table 1). In addition an unscheduled

coagulation screen including APTT, PT, TT, fibrinogen and d-dimer should be performed if there are any signs of bleeding adverse events.

4.5.4.6 O_2 saturation

A baseline measurement of oxygen saturation, performed by pulse oximetry will be recorded on Day 1 prior to dosing, at every vital sign assessment and as indicated as part of the routine management of patient on the occurrence of AEs eg, specific cardiac or respiratory symptoms. If there is a clinically significant drop in O_2 saturation this event should be followed up with full pulmonary function tests and HRCT scan of the chest if clinically indicated.

4.5.4.7 Chest X-ray

A chest X-ray will be performed at baseline and also in cases of dyspnoea and pulmonary oedema in addition to other assessments as per local standard practice. Results must be recorded in the pCRF.

Chest X-rays will be repeated as clinically indicated as part of the routine management of the patient on the occurrence of AEs.

4.5.4.8 Ophthalmological examination

A complete ophthalmologic examination including a slit-lamp examination must be performed within 2 weeks prior to first dose, and at week 5 (+/- 1 week), and if a patient experiences a visual disturbance AE. Results must be recorded in the pCRF.

4.6 Volume of blood sampling and handling of biological samples

The total volume of blood that will be drawn from each patient in this study, during their initial 35 days treatment period, is as follows:

Part A

Table 3 Part A Volume of blood to be drawn from each patient

Assessment		Sample volume (mL)	No. of samples	Total volume (mL)
Pharmacodynamic		4	6	24
Pharmacokinetic		2	32	64
Safety	Clinical chemistry	5	6	30
	BNP or NT-pro BNP	6	6	36
	Haematology	2.6	6	15.6
	Coagulation	2.7	6	16.2
Optional host genetics		9	1	9
Total				194.8

The total volumes of blood given in Table 3 is based upon a 35 days treatment period (including samples taken at the withdrawal visit). At each visit on treatment after this a further 16.3 mL (approximately) of blood will be taken for haematology, coagulation and clinical chemistry (including BNP). Clinical chemistry (including BNP, and Troponin T (or Troponin I)), coagulation and haematology samples are analysed locally, therefore volumes may vary according to local practice, and at some sites additional blood volume may be required for the Troponin T (or Troponin I) sample (approximately 2.7 mL per sample).

Table 4 – no longer applicable.

4.6.1 Analysis of biological samples

4.6.1.1 Clinical chemistry samples

All clinical chemistry samples will be analysed by the local laboratory. The analyte stability limits defined by the local laboratory will be applied to all analyses performed for this study. Samples that fall outside these stability limits should not be analysed. Analytical data will not be reported if found to have been derived from a sample that fell outside these stability limits.

4.6.1.2 Pharmacokinetic samples

The long-term stability of the analyte(s) should be documented in method validation produced by the AstraZeneca appointed CRO. Results from analyses of samples stored longer than the time period for which stability has been demonstrated should not be reported unless complementary analyte(s) stability data is acquired and appended to the relevant method validation report.

4.7 Genetic measurements and co-variables

See Appendix D and Laboratory Handbook for Investigators for details of optional blood sample collection for host genetic analysis of the study. The results of the genetic analysis will not be reported as part of the clinical study report for the study.

5. DATA MANAGEMENT

5.1 pCRF Data

Paper CRFs (pCRFs) will be provided for the recording of data not captured electronically. The pCRF will be in triplicate with carbonless paper. Data should be recorded legibly onto the forms in black ballpoint. If any data are not available, omissions will be indicated on the record forms. Corrections should be made legibly and initialled. Correction fluid or covering labels must not be used.

The AstraZeneca Monitor will check data at the monitoring visits at the investigational site. The pCRF must be signed by the Investigator to confirm that they have been checked for

accuracy and completeness; this signature should include review of all the latest test results. The top original and first copy of the completed form will be collected. The top original will be sent to data management personnel. The remaining copies will be retained at the study centre unless the Marketing Company (MC) is required to retain a copy of the CRF in their local files. In this case, the monitor will also collect the 1st copy of the CRF and retain it at the MC according to local guidance.

Data from the completed pCRFs will entered onto AstraZeneca's clinical study database and undergo validation checks for correctness and completeness under the direction of the Study Co-ordinator.

Any data queries arising from in-house validation checks, such as missing, impossible or inconsistent recordings in the pCRFs will be detailed on data query forms (DQFs) that will be forwarded to the investigator, via the monitor. After resolution of the query on the DQF, it should be photocopied twice. The original DQF will be returned to AstraZeneca. The monitor will retain a photocopy and another photocopy will be retained at the investigational site.

All data captured electronically eg, ECG results, PK results, will be transferred directly to the appropriate database. Validation and quality control of this is the responsibility of the provider.

5.2 Dose Escalation Decision Data

Dose escalation decisions will be based upon unvalidated data.

5.3 Pharmacokinetic and Pharmacodynamic Data

The pharmacokinetic and pharmacodynamic data will be fully validated before being sent to the data management technology (DMT) programmer. The data will be provided in a format agreed up front with the DMT programmer (eg, EXCEL spreadsheet).

5.4 Biomarker analysis of tumour material (Not applicable)

No longer applicable.

5.5 Pharmacogenetic Data (host)

The samples and data for host genetic analysis in this study will be coded. The samples will be labelled with a clinical study patient number that can be traced or linked back to the patient only by the investigator. The samples and data will not carry a personal identifier. The date of sample collection and confirmation that the patient has signed the appropriate informed consent form will be recorded on a pCRF.

In the case of genetic data, only the date the patient gave consent to participation in the host genetic research, the date the tumour material and blood sample was taken from the patient will be recorded in the electronic pCRF and database.

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

5.6 Reporting of genetic data (host)

Results from any host genetic research performed will be reported separately from the clinical study report. AstraZeneca will not provide individual genetic 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

The Statistical Analysis Plan (SAP) will not be prepared for this study since the detail of all planned analyses are included in this protocol.

6.2 Description of outcome variables in relation to objectives and hypotheses

The primary objective of this study is to assess the safety and tolerability of AZD8330 in patients with advanced malignancies. This objective will be assessed by AEs, clinical chemistry, haematology, urinalysis, vital signs, MUGA scan/Echocardiogram, ECGs Ophthalmologic examination, and O_2 saturation.

The secondary objective of determining the pharmacokinetics of AZD8330 and any known major metabolites following both single and multiple oral dosing of AZD8330 in patients with advanced malignancies will be assessed by derivation of pharmacokinetic parameters for both AZD8330 and any known major metabolites, which may include include C_{max}, t_{max}, AUC, CL/F, and $t_{1/2}$.

The secondary objective of investigating the extent of pERK inhibition in PBMCs, will be assessed by the change in pERK in PBMCs at 30 minutes, 1 hour 30 minutes, 4, 8 and 24 hours post-dose compared to pre-dose levels.

If the data are suitable, potential relationships between plasma AZD8330 and any known major metabolites concentrations/exposure and changes in safety parameters will be investigated using a graphical approach and/or appropriate PK/pharmacodynamic modelling techniques.

6.3 Description of analysis sets

Population	Definition
Evaluable for safety	This will include all patients who received at least one dose of study medication[a].
Evaluable for PK analysis	This is a subset of the safety population that includes all patients who provide AZD8330 concentration data and/or any known major metabolites concentration-time data.
Evaluable for Dose Escalation Evaluation (Part A)	This is a subset of the safety population that includes all Part A patients who have had all PK assessments (Day 1) performed, and who have sufficient safety evaluations performed during 35 days dose period from first dose at the discretion of the Safety Review Committee, or have suffered a DLT.

a If a patient discontinues study medication and starts another anti-cancer therapy, all data after 30-day follow-up period for that patient will be excluded from the assessment of safety.

6.4 Method of statistical analysis

No formal statistical hypothesis testing will be performed on the data from this study.

All safety, tolerability, pharmacokinetic, pharmacodynamic and efficacy data relating to each dose level will be listed and summarized as detailed below.

6.4.1 Safety Data

Safety data will not be formally analysed. All patients who receive at least one dose of AZD8330 will be included in the assessment of AE profile (evaluable for safety population). Other safety data will be assessed in terms of clinical chemistry, haematology, urinalysis, vital signs, MUGA scan/Echocardiogram, ECGs, ophthalmologic examination, and O_2 saturation. At the end of the study, appropriate summaries of all safety data will be produced, as defined below and in the SAP.

Data from all cycles of initially randomised treatment will be combined in the presentation of safety data. AEs will be listed individually by patient and dose group. For patients who have a dose modification, all AEs (due to drug or otherwise) will be assigned to the initial dose

group. The number of patients experiencing each AE will be summarised MedDRA system organ class (SOC), MedDRA preferred term (PT) and CTC grade. The number and % of patients with adverse events in different categories (e.g. causally related, CTC grade ≥3 etc) will be summarised by dose group, and events in each category will be further summarised by MedDRA SOC and PT, by dose group.

Any AE occurring before treatment (ie, before study Day 1) will be included in the data listings but will not be included in the summary tables of adverse events. In addition, a separate data listing of pre-treatment AEs will be produced.

Any AE occurring within 30 days of treatment withdrawal will be included in the AE summaries. Any events in this period that occur after a patient has received further therapy for cancer (following withdrawal of study medication) will be flagged in the data listings. AEs occurring more than 30 days after withdrawal of study medication will be listed separately, but not included in the summaries.

Clinical chemistry, haematology, urinalysis, vital signs, MUGA scan/Echocardiogram, ECGs, ophthalmologic examination, and O_2 saturations will be listed individually by patient and dose group and summarised by treatment dose, as follows:

Haematology, clinical chemistry, vital signs and ECG data will be suitably summarised by dose level for all patients by:

- summary statistics of observed values: mean, standard deviation, minimum, maximum, n
- summary statistics of change from baseline values: mean, standard deviation, minimum, maximum, n
- Individual patient listings: including highlighting of haematology and clinical chemistry values outside the local laboratory reference ranges

Qualitative assessments will be summarised for all subjects using the number of subjects with results of negative, trace or positive.

Graphical presentations of safety data will be presented as is deemed appropriate.

6.4.2 PK data

The concentration-time profiles of AZD8330 and any known major active metabolites, along with the derived pharmacokinetic variables, will be listed for each patient per dose and dosing day and summarised appropriately, as described below:

The following parameters will be reported for day 1 and day 15 if the data allow:

Summaries of AUC_{0-t}, $AUC_{0-\tau}$, C_{max}, C_{min}, CL/F and Vz/F will contain:

- the geometric mean (gmean, calculated as exp(μ), where μ is the mean of the data on a log scale)
- 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 (calculated using untransformed data)
- median, minimum, maximum
- number of observations (n).

Summaries of $t_{1/2}$ and λ_z will present:

- arithmetic mean and standard deviation
- median, minimum, maximum, n.

Summaries of t_{max} will present:

- median, minimum, maximum, n.

Summaries of plasma concentration-time data will present:

- gmean and CV
- gmean ± standard deviation (calculated as the exponential of the mean ± standard deviation on the log scale)
- arithmetic mean, standard deviation (calculated using untransformed data)
- median, minimum, maximum, n.

The following figures will be produced to present the pharmacokinetic data:

- Concentration-time profile plots for each patient (linear concentration scale), showing overlapping profiles for 0-48h for all days on which PK samples are available as separate lines.
- Concentration-time profile plots for each patient (log concentration scale), showing overlapping profiles for 0-48h for all days on which PK samples are available as separate lines.
- Concentration-time profile plots for each patient (linear concentration scale), showing a single profile for the entire dosing period.

- Concentration-time profile plots for each patient (log concentration scale), showing a single profile for the entire dosing period.

- Gmean +/- standard deviation (one line for each dose level, linear concentration scale)

- Gmean +/- standard deviation (one line for each dose level, log concentration scale)

- Dose-normalised C_{max} and AUC versus dose

Additional or alternative plots may be constructed as deemed appropriate based on emerging data.

Non-quantifiable (NQ) values of plasma concentrations will be handled as follows:

- if, at a given time point, 50% or less of the plasma concentrations are non-quantifiable (NQ), the gmean, CV, gmean ± standard deviation, arithmetic mean and standard deviation will be calculated by substituting the limit of quantification (LOQ) for values which are NQ.

- if more than 50%, but not all, of the concentrations are NQ, the gmean, CV, gmean ± standard deviation, arithmetic mean and standard deviation will be reported as not calculable (NC)

- if all the concentrations are NQ, the gmean and arithmetic mean will be reported as NQ and the CV, gmean ± standard deviation and standard deviation as NC

- if the calculation of the gmean – standard deviation results in a value less than the LOQ, NQ will be displayed.

6.4.3 Pharmacodynamic data

6.4.3.1 PBMC biomarkers

The levels of biomarker pERK in PBMCs, and their baseline scaled ratios (BSRs), defined as post-dose ÷ pre-dose, will be listed and summarised by dose group.

If the data are suitable the extent of pERK inhibition in PBMCs following the oral doses in this study will be investigated in all patients with pre and post dose samples using graphical means, including scatter plots of the following for each biomarker:

- BSR vs dose group

- BSR vs pre-dose biomarker level

6.4.3.2 Tumour response

Summaries of best overall response and Objective Response Rate by initial treatment will be produced, for all patients from the safety population who have a baseline RECIST assessment and at least one follow-up RECIST assessment.

Graphical presentations including the following will be produced:

Waterfall plots for each dose level showing the sum of the changes in target lesions for each patient at week 8 compared to baseline, indicating tumour type for each patient.

Scatter plots of change in lesion length vs pre-dose biomarker levels

Scatter plots of change in lesion length vs biomarker BSR

6.4.4 PK/pharmacodynamic relationships

If the data are suitable, the relationship between the plasma AZD8330 and any known major metabolites concentrations/exposure and efficacy/safety parameters will be investigated either using graphical means or appropriate PK/PD software. Details of the pharmacokinetic analyses will be pre-defined prior to data lock.

6.5 Determination of sample size

Part A of the study is not formally powered but is designed to provide adequate tolerability, safety, pharmacokinetic, and pharmacodynamic data.

Part A is also designed to ensure at least 6 evaluable patients are recruited at the dose deemed to be MTD.

The number of patients has been based on both feasibility and obtaining adequate data to assess the biological activity of AZD8330 in tumour in terms of the biomarker pERK in tumour, based on average with-patient reduction in pERK from baseline, as measured using semi-quantitative IHC methods.

Average reductions of 83% on pERK were observed in the first in man study with the MEK inhibitor AZD6244 (Adjei A et al 2006). The within patient standard deviation of the change from baseline (on the log scale) from this study is estimated as 2.2. 10 patients would therefore provide 80% power to detect an average 70% reduction at the 1-sided 20% significance level.

Therefore approximately 10 evaluable patients at each dose level are considered reasonable to indicate the possibility of biological activity.

The expansion also provides an opportunity to provide a better estimate of the tolerability of each dose for onward development. Expanding the cohort at the maximum tolerated dose from the dose escalation to around 20 provides a stronger estimate of the true DLT rate. For example if the MTD cohort had only 6 patients then:

- If 0 out of 6 DLTs are observed it can be concluded that the true DLT rate is less than 30% with 90% confidence.

- If 1 DLT (16.7%) is observed it can be concluded that the true DLT rate is less than 50% with 90% confidence.

However, with an additional 18 evaluable patients for example:

- If 0 out of 24 DLTs are observed it can be concluded that the true DLT rate is less than 10% with 90% confidence.

- If 4 out of 24 DLTs (16.7%) are observed it can be concluded that the true DLT rate is less than 32% with 90% confidence.

6.6 Interim analyses

No formal statistical interim analysis will be undertaken.

6.7 Data monitoring board (Not applicable)

Not applicable to this Study.

6.8 Study Safety Review Committee

A study Safety Review Committee will meet during Part A following each dose step (ie, following one cycle of treatment for each dose level). The Safety Review Committee comprising of the Investigators, the Medical Science Director, the Study Team Physician, the Pharmacokineticist and the Global Drug Safety Physician (or nominated deputy in each case) will assess the available safety, tolerability and PK data. The committee must include a minimum of 3 physicians at least 1 of who must be an investigator, but may also include other team members, if appropriate (eg, statistician etc).

Further details of the Safety Review Committee can be found in the Safety Review Committee Guidelines.

7. STUDY MANAGEMENT

7.1 Monitoring

Before first subject 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 pCRFs, and that investigational product accountability checks are being performed.
- Perform source data verification (a comparison of the data in the pCRFs with the subject'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 subject (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 subject 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 genetic analysis 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 International 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 at each centre 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 subject'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 overall timetable for the study is summarised below:

Study period		**Phase of development**
Estimated date of first patient enrolled	April 2007	Phase I
Estimated date of last patient completed	August 2010*	

* End of study is defined as the point at which the last patient has received 6 months of treatment with AZD8330, or has withdrawn from the study (whichever is sooner).

In Part A of this study, patients may continue to receive AZD8330 until disease progression or as long as they continue to derive benefit from treatment. Therefore, the time at which the last patient stops treatment cannot be precisely predetermined. The study endpoint is defined as the point at which the last patient has received 6 months of treatment with AZD8330, or has withdrawn from the study (whichever is sooner). The study database will be locked at this point and the Clinical Study Report will be written. Serious adverse event reports for any patients still receiving treatment after the end of study will be entered on the AstraZeneca Global Patient Safety database. The study database will not be updated with this information nor an addendum to the Clinical Study Report be written.

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 subject 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 subject participates in this genetic research.

For US only: The Principal Investigator is also responsible for providing the Institutional Review Board with reports of any serious and unexpected adverse drug reactions from any other study conducted with the investigational product. AstraZeneca will provide this information to the Principal Investigator.

Progress reports and notifications of serious and unexpected adverse drug reactions will be provided to the Ethics Committee according to local regulations and guidelines.

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 4.7.

8.3 Informed consent

The principal investigator(s) at each centre will ensure that the subject 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 subject should be given the opportunity to ask questions and allowed time to consider the information provided.

The subject's signed and dated informed consent must be obtained before conducting any procedure specifically for the study, including the following:

- Collection of study blood samples
- Study ECGs
- Study Echocardiograms/MUGA scans
- Ophthalmological examination

Host Genetics Research will be covered separately in the informed consent forms, and all data protection and confidentiality principles, described in the main study protocol, are also applicable to the genetic research.

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

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 / study code / initials.

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.

All data protection and confidentiality principles, described in the main study protocol, are also applicable to the genetic research.

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 genetic 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 subject, however, it must be recognised that there are exceptional circumstances where individuals may see both genetic data and a subject'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 you may contact the Study Delivery Team Physician. If the Study Delivery Team Physician is not available, contact the Study Delivery Team Leader at the AstraZeneca Research and Development site shown below.

Role in the study	Name	Address & telephone number

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 4.5.1.

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 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 pCRFs.
- An overdose with associated non-serious AEs should be recorded as the AE diagnosis/symptoms on the relevant AE forms in the 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 pCRFs. The overdose should be reported on the separate AZ "Clinical Study Overdose Report Form".

9.4 Procedures in case of pregnancy

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 and documented even if the subject was discontinued from the study. Should a patient become pregnant they should immediately be withdrawn from study treatment.

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 pregnancies, following maternal or paternal exposure, should be reported to AstraZeneca and the outcome (spontaneous miscarriage, elective termination, normal birth or congenital abnormality) followed up, even if a patient has discontinued from the study. Pregnancies and outcomes should be reported using a Pregnancy Outcome Report (Part I & II).

10. REFERENCES

Adjei A et al 2006
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Clinical Study Protocol: Appendix B

Drug Substance	AZD8330
Study Code	D1536C00001
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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.

Examples of such events are:

- Angioedema not severe enough to require intubation but requiring iv hydrocortisone treatment
- Hepatotoxicity caused by paracetamol (acetaminophen) overdose requiring treatment with N-acetylcysteine
- Intensive treatment in an emergency room or at home for allergic bronchospasm
- Blood dyscrasias (eg, neutropenia or anaemia requiring blood transfusion, etc) or convulsions that do not result in hospitalisation
- Development of drug dependency or drug abuse.

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	AZD8330
Study Code	D1536C00001
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Appendix Date	

Appendix C
Definitions of measurable, non-measurable, target and non-target lesions and treatment evaluation response based on the RECIST (Response Evaluation Criteria in Solid Tumours) criteria (Therasse et al, 2000)

1. INTRODUCTION

This appendix details the implementation of RECIST for the D1536C00001 study in regards to the investigator site review, including modifications specific for this study.

2. SCHEDULE OF EVALUATION

All baseline evaluations should be performed as closely as possible to the beginning of treatment and never more than 4 weeks before the beginning of treatment. Baseline assessment must be able to adequately define all areas of disease.

The same method of assessment and the same technique should be used as at baseline.

Any other sites at which new disease is suspected should also be imaged at follow up.

All imaging should be performed according to the study plan (see protocol Table 1)

DEFINITION OF MEASURABLE AND NON-MEASURABLE LESIONS

Measurable:

- Lesions that can be accurately measured in at least one dimension (longest diameter to be recorded) as ≥ 20 mm with conventional techniques or as ≥ 10 mm with spiral CT scan

Non-measurable:

- All other lesions, including small lesions (longest diameter < 20 mm with conventional techniques or < 10 mm with spiral CT scan)

- Truly non-measurable lesions include the following: bone lesions; leptomeningeal disease; ascites; pleural / pericardial effusion; inflammatory breast disease; lymphangitis cutis/pulmonis; abdominal masses that are not confirmed and followed by imaging techniques; cystic lesions

3. METHODS OF MEASUREMENT

The same method of assessment and the same technique should be used to characterize each identified and reported lesion at baseline and during follow-up.

All measurements should be recorded in metric notation by use of a ruler or calipers.

Imaging based evaluation is preferred to evaluation by clinical examination when both methods have been used to assess the anti-tumour effect of a treatment.

3.1 Clinical examination (non target lesions only)

Clinical lesions will only be considered measurable when they are superficial (eg, skin nodules, palpable lymph nodes). For the case of skin lesions, documentation by color photography including a ruler to estimate the size of the lesion is recommended.

In the D1536C00001 study clinical assessment will not be used for measuring target lesion selected for response assessment. This method of assessment will be used in this study for non-targets or new lesions.

3.2 Chest x-ray (not used in this study)

According to RECIST, lesions on chest X-ray are acceptable as measurable lesions when they are clearly defined and surrounded by aerated lung. However, CT is preferable.

In this study, chest x-ray will not be used as part of RECIST as CT and MRI are the best currently available and most reproducible methods for measuring target lesions selected for response assessment.

Note: Plain x-ray may only be used for assessing bone lesions as non target lesions or confirming a new lesion on bone scan.

3.3 CT and MRI (target and non target lesions)

CT and MRI are generally considered to be the best currently available and reproducible methods to measure target lesions selected for response assessment. Conventional CT and MRI should be performed with cuts of 10 mm or less in slice thickness contiguously. Spiral CT should be performed using a 5 mm contiguous reconstruction algorithm. This applies to the chest, abdomen and pelvis. Head & neck and extremities usually require specific protocols.

In the D1536C00001 (AZD8330) study, it is recommended that baseline CT examination to be performed to adequately define all areas of disease. Post-baseline imaging should follow and evaluate all previous identified lesions. MRI should only be used where CT is not feasible or it is medically contra-indicated.

3.4 Ultrasound (not used in this study)

Ultrasound (US) should not be used to measure tumour lesions for objective response evaluation. It is however a possible alternative to clinical measurements of superficial palpable nodes, subcutaneous lesions and thyroid nodules. US might also be useful to confirm the complete disappearance of superficial lesions usually assessed by clinical examination.

As ultrasound is not appropriate for assessing objective response, it will not be used as part of the RECIST assessment in the D1536C0001 study.

3.5 Endoscopy and laparoscopy (not used in this study)

The utilization of these techniques for objective tumour evaluation has not yet been fully and widely validated. Their uses in this specific context require sophisticated equipment and a high level of expertise that may only be available in some centers. However, such techniques can be useful to confirm complete pathological response when biopsies are obtained.

These methods will not be used as part of the RECIST assessment in this study.

As endoscopy and laparoscopy are not appropriate for assessing objective response, it will not be used as part of the RECIST assessment in the D1536C00001 study.

3.6 Tumour markers (not used in this study)

According to RECIST criteria, tumour markers alone cannot be used to assess response. If markers are initially above the upper normal limit, they must normalize for a subject to be considered in complete clinical response.

Tumour markers will not contribute to the response assessment in D1536C00001 study.

3.7 Cytology and histology

These techniques can be used to differentiate between PR and CR in rare cases (for example, residual lesions in tumour types such as germ cell tumours, where known residual benign tumours can remain). The cytological confirmation of the neoplastic origin of any effusion that appears or worsens during treatment when the measurable tumour has met criteria for response or stable disease is mandatory to differentiate between response or stable disease (an effusion may be a side effect of the treatment) and progressive disease.

In the absence of negative cytology findings for pleural effusion that worsens or appears, this will be considered to be disease progression due to new lesions or progression of non-target lesions.

Note: Radioisotope bone scans will not be used to assess bone lesions as non-target or new lesions as it is not an accepted method of assessment for RECIST. Bone lesions will be assessed using x-ray, CT or MRI and recorded as non-target or new lesions.

4. TUMOUR RESPONSE EVALUATION

4.1 Assessment of overall tumour burden and measurable disease

To assess the objective response during the study, it is necessary to estimate the overall tumour burden at baseline to which subsequent measurements will be compared.

In this study patients with at least one measurable site of disease as defined by RECIST criteria will be included.

Measurable disease is defined by the presence of at least one measurable lesion.

If the measurable disease is restricted to a solitary lesion, its neoplastic nature should be confirmed by cytology/histology.

4.2 Target lesions

4.2.1 Documentation of target lesions

All measurable lesions up to a maximum of 10 lesions representative of all involved organs (maximum of 5 lesions per organ) should be identified as target lesions and will be recorded and measured at baseline.

Target lesions should be selected on the basis of their size (lesions with the longest diameter) and their suitability for accurate repetitive measurements (either by imaging techniques or clinically).

A sum of the longest diameter (LD) for all target lesions will be calculated and reported as the baseline sum LD. The baseline sum LD will be used as reference to further characterize the objective tumour response of the measurable dimension of the disease.

The longest diameter will be measured and recorded for all target lesions identified at baseline at follow-up assessments and the sum LD calculated. For lesions measurable in 2 or 3 dimensions, always report the longest diameter at the time of each assessment.

If a lesion splits into two or more parts, then the sum of the LDs of those parts is recorded.

If two or more lesions merge, then the LD of the combined lesion should be recorded for one of the lesions and zero recorded for the other lesion.

If a lesion becomes to small to measure, then the size below which measurement cannot be accurately obtained should be substituted for the LD and used in the sum LD.

If a lesion cannot be measured accurately due to progression, then the maximum measurable LD should be used in the sum LD and response assessment.

If a lesion cannot be measured accurately due to it being too large, and was measurable previously, then the maximum measurable size should be recorded as the LD and should be used in the sum LD and response assessment.

4.2.2 Evaluation of target lesions:

This section provides the definitions of the criteria used to determine objective tumour response for target lesions:

Complete Response (CR)	Disappearance of all target lesions
Partial Response (PR)	At least a 30% decrease in the sum of LD of target lesions taking as reference the baseline sum LD
Progressive Disease (PD)	At least a 20% increase in the sum of LD of target lesions taking as references the smallest sum LD recorded (either at baseline or at previous assessment since treatment began)
Stable Disease (SD)	Neither sufficient shrinkage to qualify for PR nor sufficient increase to qualify for PD

Note: Appearance of new lesions only counts towards the overall visit response, not towards the response of target or non-target lesions

4.3 Non-Target lesions

4.3.1 Documentation of non- target lesions

All other lesions (or sites of disease) not recorded as target lesions should be identified as non-target lesions and should also be recorded at baseline.

Measurements are not required for these lesions, but these should be followed as "present", "absent" or "present with progression" at subsequent visits.

Note: These criteria will be used to assess all patients in this study with non target lesions

4.3.2 Evaluation of non-target lesions

This section provides the definitions of the criteria used to determine objective tumour response for non- target lesions:

Complete Response (CR)	Disappearance of all non-target lesions
Non-Complete Response / Non-Progression (Non CR-Non PD)	Persistence of one or more non-target lesion or/and maintenance of tumour marker level above the normal limits.
Progression (PD)	Unequivocal progression of existing non-target lesions.

Notes:

- Patients with a global deterioration of health status requiring discontinuation of treatment without objective evidence of disease progression at that time should be classified as having “symptomatic deterioration”. Every effort should be made to document the objective disease progression, even after discontinuation of treatment.

- In some circumstances, it may be difficult to distinguish residual disease from normal tissue. When the evaluation of complete response depends on this determination, it is

recommended that the residual lesion be investigated (fine-needle aspiration/biopsy) before confirming the complete response status.

4.4 Evaluation of Best Overall Response

The best overall response is the best response recorded from the start of treatment until disease progression/recurrence (taking as reference for progressive disease the smallest measurements recorded since the treatment started).

In general, the patient's best response assignment will depend on the achievement of both measurement and confirmation criteria.

Target lesions	Non-target lesions	New lesions	Overall response
CR	CR	No	CR
CR	Incomplete response/SD	No	PR
PR	Non-PD	No	PR
SD	Non-PD	No	SD
PD	Any	Yes or No	PD
Any	PD	Yes or No	PD
Any	Any	Yes	PD

CR = complete response; PR = partial response; SD = stable disease; PD = progressive disease

For each visit where tumour response evaluation assessments are performed, the sponsor will derive the overall visit response, using the algorithm shown above, based on the response of the target lesions, the non-target lesions and the presence or absence of new lesions for patients with measurable disease at baseline.

4.4.1 Frequency of tumour re-evaluation

Frequency of tumour re-evaluation while on treatment should be protocol specific and adapted to the type and schedule of treatment.

Frequency of tumour evaluation while on treatment for the D1536C00001 AZD8330 study it is describe in Study plan Table 1 in the Study Protocol. After baseline evaluation a tumour assessments should be perform at every 8 weeks.

5. CONFIRMATORY MEASUREMENT

5.1 Confirmation

To be assigned a status of PR or CR, changes in tumour measurements must be confirmed by repeat assessment that should be performed no less than 4 weeks after the criteria for response were first met.

In this study, the confirmation of response (CR or PR) it is determined by study protocol to be performed at next schedule visit.

In the case of stable disease, measurements must have met the stable disease criteria at least once after study entry at a minimum interval (in general, not less than 6–8 weeks).

For D1536C00001 study the interval defined in the study protocol is 8 weeks.

5.2 Duration of overall response

The duration of overall response is measured from the time that measurement criteria are met for complete response or partial response (whichever status is recorded first) until the first date that recurrent or progressive disease is objectively documented, taking as reference for progressive disease the smallest measurements recorded since the treatment started.

The duration of overall complete response is measured from the time measurement criteria are first met for complete response until the first date that recurrent disease is objectively documented.

5.3 Duration of stable disease

Stable disease is measured from the start of the treatment until the criteria for disease progression is met, taking as reference the smallest measurements recorded since the treatment started.

6. SPECIFICATIONS FOR RADIOLOGICAL IMAGING

These notes are recommendations for use in clinical studies and as such these protocols for computed tomography (CT) and magnetic resonance imaging (MRI) scanning may differ from those employed in clinical practice at various institutions. The use of standardized protocols allows comparability both within and between different studies, irrespective of where the examination has been undertaken.

CT

CT scans of the thorax, abdomen and pelvis should be contiguous throughout the anatomical region of interest. The type of CT scanner is important regarding the slice thickness and minimum sized lesion. For spiral (helical) CT scanners, the minimum size of any given lesion at baseline may be 10 mm, provided the images are reconstructed contiguously at 5mm

intervals. For conventional CT scanners, the minimum sized lesion should be 20 mm using a contiguous slice thickness of 10 mm.

Other body parts, where CT scans are of different slice thickness, (such as the neck, which are typically of 5 mm thickness) or in the young pediatric population, where the slice thickness may be different, the minimum sized lesion allowable will be different.

In subjects in whom the abdomen and pelvis have been imaged, oral contrast agents should be given to accentuate the bowel from other soft tissue masses.

Intra-venous (IV) contrast agents should also be given, unless contra-indicated for medical reasons, such as allergy. This is to accentuate vascular structures from adjacent lymph node masses and to help enhance liver and other visceral metastases. The method of administration of IV contrast agents is variable. It is appropriate to suggest that an adequate volume of a suitable contrast agent should be given such that the metastases are demonstrated to best effect and a consistent method is used on subsequent examinations for any given subject.

All images from each examination should be included and not "selected" images of the apparent lesion.

All window settings should be included, particularly in the thorax where lung and soft tissue windows should be considered. When measuring lesions, lesions should be measured on the same window setting on each examination. It is not acceptable to measure a lesion on lung windows on one examination, then on soft tissue settings on the next. In the lung, it does not really matter whether lung or soft tissue windows are used for intra-parenchymal lesions, provided a thorough assessment of nodal and parenchymal disease has been undertaken and the target lesions are measured as appropriate using the same window settings for repeated examinations throughout the study.

MRI

MRI is a complex issue. MRI is entirely acceptable and capable of providing images in different anatomical planes. It is important therefore that when it is used lesions must be measured in the same anatomical plane using the same imaging sequences on subsequent examinations. MRI scanners vary in the images produced. Wherever possible, the same scanner should be used. Moreover many subjects with advanced malignancy are in pain, so their ability to remain still for the duration of a scan sequence, in the order of 2-5 minutes is limited. Any movement during the scan time leads to motion artifacts, degradation of image quality such that the examination will probably be useless.

For these reasons, CT is at this point in time the imaging modality of choice.

Same method

The same imaging modality must be used throughout the study to measure disease. Different imaging techniques have differing sensitivities, so any given lesion may have different dimensions at any given time if measured with different modalities. It is therefore, not

acceptable to interchange different modalities throughout a trial and use these measurements. It must be the same technique throughout.

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Clinical Study Protocol Appendix D	
Drug Substance	AZD8330
Study Code	D1536C00001
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Appendix D
Optional Genetic Research

GENETICS RESEARCH SYNOPSIS

A Phase I, Open-Label, Multi-centre Study to Assess the Safety, Tolerability and Pharmacokinetics of Single and Multiple Oral Doses of AZD8330 in Patients With Advanced Malignancies

The genetic research activities described in this appendix (including the collection and storage of genetic samples), are optional for study sites as well as for individual patients. These research activities will hereafter be referred to as "this genetic research." The clinical study protocol to which this document is appended will be referred to as "the main study". The term "genetic sample" means a blood sample collected for genetic research and/or DNA prepared from it.

This genetic research will be performed only after the appropriate Ethics Committee has approved it. Informed consent will be obtained using a form separate from that used for the main study. All sections of the protocol for the main study also apply to this genetic research.

Study centre(s) and number of patients who may be enrolled in this genetic research

As per main protocol.

Objectives

Exploratory Objectives	Outcome variables
To collect a blood sample (optional) for DNA extraction and storage to investigate whether variability in the AZD8330 PK, safety, efficacy or pharmacodynamic results could be explained by differences in the patient's genotype.	Correlation of host polymorphisms with variation in PK, safety or response parameters observed in patients treated with AZD8330

Study design

It is proposed to collect an optional blood samples for genetic analysis. Provision of a blood sample for genetic analysis will be optional for all patients entering the study and will involve a separate consent procedure. A patient's acceptance of this procedure will not be a requirement for his or her participation in the main study.

The blood sample and data for pharmacogenetic analysis in this study will be coded. Each sample will be labelled with the study number and patient enrolment number (E-code). Only the investigator will be able to link the sample to the individual patient. The sample and data will not be labelled with a personal identifier.

Target population

All consenting patients in all centres participating in the main part of this study

Statistical methods

The number of patients who will agree to participate in this genetic research is unknown. It is therefore not possible to establish whether sufficient data will be generated. A statistical analysis plan will be prepared where appropriate.

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LIST OF ABBREVIATIONS AND DEFINITION OF TERMS

Abbreviation or special term	Explanation
°C	Degrees Celsius
ADME	Absorption/Distribution/Metabolism and Excretion
AE	Adverse Event
CRF	Case record form
CSR	Clinical study report
DNA	Deoxyribonucleic acid
EDTA	Ethylenediamine tetra-acetic acid
ICH	International Conference on Harmonisation
LIMS	Laboratory information management system
PD	Pharmacodynamic
PK	Pharmacokinetic
mL	Millilitre
UK	United Kingdom

1. BACKGROUND

AstraZeneca plan to include investigations into genetic variations and their effect on drug response as part of the drug development program for all projects where it is considered to be appropriate. By using this information, the aim is to better understand the impact of genetic variation and how it can be utilised to bring better drugs to the market.

To achieve this goal a systematic collection of deoxyribonucleic acid (DNA) for genetic analysis (derived from blood samples taken from consenting study patients) will be implemented across a broad range of relevant clinical studies. The ability to acquire appropriate consent to collect blood samples to establish an archive and allow future meta-analysis of data derived from a number of studies for AZD8330 is of the utmost importance. This genetic research forms part of this strategy.

1.1 Rationale for genetic research

AstraZeneca intends to perform genetic research in the AZD8330 clinical development programme to explore how genetic variations may affect the clinical parameters associated with AZD8330.

The benefits of being able to explore associations between genes and clinical outcomes within the AZD8330 programme are potentially many including the possibility to explain potential outliers, such as non-responders or to explain potential adverse reactions related to drug exposure.

2. GENETIC RESEARCH OBJECTIVES

Genes that may be investigated include those which may be of relevance to the Absorption/ Distribution/Metabolism/Excretion (ADME) of AZD8330. Additional information on other genes important for this drug and for the response to AZD8330 in cancers for which the drug is being developed will become available in the future and it is, therefore important to retain the possibility of investigating additional genes in the context of AZD8330 clinical program.

3. GENETIC RESEARCH PLAN AND PROCEDURES

3.1 Genetic Research plan

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

The patient will be asked to participate in this genetic research during their enrolment or screening visit.

If the patient agrees to participate a 9 mL blood sample will be collected into a tube containing ethylenediamine tetra-acetic acid (EDTA). Blood samples will ideally be collected prior to dosing with AZD8330. Although genotype is a stable parameter, early sample collection is preferred to avoid introducing bias through excluding patients who may withdraw due to an adverse event (AE), such patients would be important to include in any genetic analysis. If for any reason the sample is not drawn at enrolment, it may be taken at any visit until the last study visit.

3.2 Selection of genetic research population

3.2.1 Study selection record

All patients who take part in the study will be asked to participate in this genetic research. Participation is voluntary and if a patient declines to participate there will be no penalty or loss of benefit. The patient will not be excluded from any aspect of the main study.

3.2.2 Inclusion criteria

For inclusion in this genetic research, patients must fulfil all of the inclusion criteria described in the main body of the study protocol **and**:

- Provide informed consent for the genetic sampling and analyses.

3.2.3 Exclusion criteria

Exclusion from this genetic research may be for any of the exclusion criteria specified in the main study or any of the following:

- Previous allogenic bone marrow transplant
- Blood transfusion in the last 120 days prior to pre-entry visit

3.2.4 Discontinuation of patients from this genetic research

3.2.4.1 Criteria for discontinuation

Specific reasons for discontinuing a patient from this genetic are:

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

3.2.4.2 Procedures for discontinuation

Patients who discontinue from the main 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(s) being kept for genetic research in the future

- Withdraws consent for the samples to be kept for genetic research in the future and wishes the samples to be destroyed. Destruction of the samples (or the DNA extracted 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.

4. GENETIC MEASUREMENTS AND CO-VARIABLES

4.1 Summary of genetic objectives and analysis

The purpose of this genetic research is to generate data for use in future retrospective or prospective analyses. Future analyses will explore genetic factors, which may influence the disposition, efficacy, safety and tolerability to AZD8330 and/or susceptibility to or prognosis of cancer. The results of this 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 AZD8330 to generate hypotheses to be tested in future studies.

4.2 Collection of samples for genetic research

Patients will provide blood sample(s) as per the inclusion criteria and visit schedule.

A 9 mL blood sample will be collected into a polypropylene tube containing ethylenediamine tetra-acetic acid (EDTA) and gently inverted a minimum of five times to mix thoroughly. The sample will then be stored at -20°C prior to shipment. Tubes will identifiable for 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 the CRF.

Genotype is a stable parameter, therefore if for any reason the blood sample is not drawn at their enrolment or screening visit, 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.

4.2.1 Sample processing and shipping

AstraZeneca or it's designee will act as the central laboratory for sample logistics. This will include the supply of site material and all transport arrangements.

A single blood sample will be stored frozen (-20°C or below) at the site and sent to the central laboratory. The central laboratory will then send the samples to AstraZeneca or it's designee laboratory for DNA extraction. Samples must remain frozen at all times. Further details on the processing of the samples are outlined in the Laboratory Manual for Investigators.

Where possible samples should be shipped in batches and shipment will be co-ordinated with the receiving site to ensure samples arrive within working hours, on normal working days. Details of protocol study number, centre number, patient e-code, and/or randomisation number and date of sample collection should accompany the shipment.

4.2.2 Storage and coding of DNA samples

The following procedures refer to the processed adopted for the coding and storage of the samples for genetic analysis. 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 be 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 identified with the study number and patient enrolment number. Only the investigator will be able to link the blood sample to the individual patient. The sample and data will not be labelled with a personal identifier. The link between the patient enrolment/randomisation code and the DNA number will be maintained.

This link file and any corresponding genetic data will be stored in a secure environment, with restricted access within the Clinical Genotyping Group Laboratory Information Management System (LIMS) or other appropriate system at AstraZeneca, Alderley Park, UK. The link will be used to identify the relevant DNA samples for analysis, facilitate correlation of genotypic results with clinical data, allow regulatory audit and to trace samples for destruction in the case of withdrawal of consent. Access to the link file will require written authorisation from the Clinical Development Team Leader.

All DNA samples will be stored under secure conditions with restricted access at AstraZeneca or the contracted laboratory. The blood and 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.

5. MANAGEMENT OF GENETIC RESEARCH DATA

Genetic data, derived from blood or DNA will not be reported in the CSR.

In the case of genetic data, only the date the patient gave consent to participation in the genetic research and the date the blood sample was taken from the patient will be recorded in the CRF and database. The genetic data will not be merged with the clinical dataset, residing within the clinical database, collected from the patient population for statistical analysis.

The genetic 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 from 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 Reporting of genetic results

Results from this genetic research will be reported separately from the clinical study report for the main study. 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

The number of patients who will agree to participate in the genetic research is unknown. It is therefore not possible to establish whether a statistically relevant number of patients will consent to provide sufficient data to 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.

7. STUDY MANAGEMENT

7.1 Monitoring

Before first patient entry into the study, a representative of AstraZeneca will visit the investigational study site. In addition to the requirements described in the main study, this genetic research will be discussed.

During the study, a representative of AstraZeneca will have regular contacts with the investigational site. One of the purposes of these visits will be to perform source verification of the genetic consent of participating patients and to ensure that the investigational team are adhering to the specific requirements of this genetic research.

7.2 Training of staff

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 a representative of AstraZeneca. The ethical considerations specific to genetic analysis and the importance of the informed consent process will be made clear. The requirements for the collections of the patients' sample will also be made clear.

7.3 Changes to the protocol

Any changes to the genetic research will comply with the principles described in Section 7.4 of the main body of the protocol.

7.4 Study agreements

The principal investigator at each centre 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. Specific reference to requirements relating to this genetic research will be included in the study agreement(s).

8. ETHICS

8.1 Ethics review

In addition to documenting Ethics Committee approval of the main study, approval must be obtained for this genetic research and the associated genetic informed consent from the relevant 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 4.2.2 of this Appendix.

8.3 Informed consent

The genetic component of this study is optional and the patient may participate in other components of the study without participating in the genetic component. To participate in the genetic component of the study which involves donation of blood sample, the patient must sign and date both the consent form for the main study and the consent form for 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 from the genetic aspect of the study at any time.

8.4 Patient data protection

All data protection and confidentiality principles, described in the main study protocol, are applicable to this genetic research.

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 genetic 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 patients 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. REFERENCES

None

Clinical Study Protocol Appendix E	
Drug Substance	AZD8330
Study Code	D1536C00001
Edition Number	1
Date	

Appendix E
Cockcroft-Gault formula

The Cockcroft-Gault formula has been provided, as the protocol allows for the serum creatinine to be calculated using the Cockcroft-Gault formula (see Section 3.3.2, Inclusion criteria):

For serum creatinine values in mol/L:

Men: [(140 – age) x weight (kg) x 1.23] / creatinine (μmol/L)

Women: [(140 – age) x weight (kg) x 1.04] / creatinine (μmol/L)

For serum creatinine values in mg/dL:

Men: [140 – age] x weight (kg) / [72 x creatinine (mg/dL)]

Women: 0.85 x [140 – age] x weight (kg) / [72 x creatinine (mg/dL)]

Ref: Cockcroft D, Gault MD. Nephron 16: 31- 41, 1976.