FOCUS4 – Molecular selection of therapy in colorectal cancer: a molecularly stratified randomised controlled trial programme

FOCUS4-C Protocol

A randomised controlled comparison of AZD1775 (WEE1 inhibitor) versus active monitoring in patients with colorectal cancer that is stable or responding after first-line treatment in two biomarker groups:

- Patients with H3K36me3 loss tumours
  OR
- Patients with tumours mutated at both RAS and P53

Version: 5.0
Date: 05\textsuperscript{th} April 2018
MRC CTU ID: CR13
ISRCTN #: ISRCTN90061546
EUDRACT #: 2012-005111-12
CTA #: 00316/0245/001-0001
REC #: 13/SC/0111

Authorised by:
Name: Professor Matt Seymour/Dr Jenny Seligmann
Role: FOCUS4-C Co-Chief Investigators
Signature:
Date: 25\textsuperscript{th} April 2018 / 19\textsuperscript{th} April 2018

Name: Professor Tim Maughan
Role: FOCUS4 Trial Programme Chief Investigator
Signature:
Date: 11\textsuperscript{th} April 2018

Name: Professor Richard Kaplan
Role: FOCUS4 MRC Programme Lead
Signature:
Date: 17\textsuperscript{th} April 2018
GENERAL INFORMATION

This document was constructed using the Medical Research Council (MRC) Clinical Trials Unit (CTU) at University College London (UCL) Protocol Template Version 4.0. In combination with the FOCUS4 Master Protocol, it describes the overall plan and structure for FOCUS4-C. The FOCUS4 Trial Programme is coordinated by the MRC CTU at UCL, and this document provides information about procedures for entering patients into FOCUS4-C after a patient has been registered through the procedures described in the FOCUS4 Master Protocol.

The FOCUS4 Trial Programme protocols should not be used as aide-memoire or guide for the treatment of other patients. Every care has been taken in drafting the protocols, but corrections or amendments may be necessary. These will be circulated to the registered investigators in the trials, but sites entering patients for the first time are advised to contact the FOCUS4 Trial team at the MRC CTU at UCL, London, UK, to confirm they have the most up-to-date versions.

COMPLIANCE

The FOCUS4 Trial Programme will be conducted in compliance with the approved protocol, the 1996 version of the Declaration of Helsinki, the principles of Good Clinical Practice (GCP), EU Directives 2001/20/EC Article 2 and 2005/28/EC and subsequent amendments, their implementation in national legislation in the UK by Statutory Instrument 2004/1031 and subsequent amendments, the UK Data Protection Act (DPA number: Z6364106), and the National Health Service (NHS) Research Governance Framework for Health and Social Care (RGF).

SPONSOR

The MRC is the trial sponsor for the FOCUS4 Trial Programme and has delegated responsibility for the overall management of the FOCUS4 Trial Programme to the MRC CTU at UCL. Queries relating to MRC sponsorship of this trial should be addressed to the Director, Professor Max Parmar, Institute of Clinical Trials & Methodology, MRC CTU at UCL, 90 High Holborn 2nd Floor, London WC1V 6LJ UK or via the FOCUS4 trial team.

FUNDING

The FOCUS4 Trial Programme is jointly funded by the MRC/NIHR Efficacy and Mechanism Evaluation (EME) programme and Cancer Research UK (CRUK). Funding for drug and distribution (via Fisher Clinical Services) is provided by AstraZeneca in the UK.

AUTHORISATIONS AND APPROVALS

This FOCUS4 Trial Programme and all current comparisons within it were approved by the Oxford South Central – Panel C Ethics Committee and Medicines and Healthcare Products Regulatory Agency and is part of the NIHR CRN Portfolio.

TRIAL REGISTRATION AND RANDOMISATION

This FOCUS4 Trial Programme has been registered with the ISRCTN Clinical Trials Register, where it is identified as ISRCTN90061546.
FOCUS4-C ADMINISTRATION

FOCUS4-C CO-CHIEF INVESTIGATORS
Professor Tim Maughan  Email: tim.maughan@oncology.ox.ac.uk
Professor Matt Seymour  Email: matt.seymour@nihr.ac.uk
Dr Jenny Seligmann  Email: J.Seligmann@leeds.ac.uk

MRC CO-ORDINATING CENTRE
Institute of Clinical Trials & Methodology
MRC Clinical Trials Unit at UCL (MRC CTU)  Fax: 020 7670 4653
90 High Holborn  SAE Fax: 020 7670 4818
2nd Floor  Email: mrcctu.focus4@ucl.ac.uk
London WC1V 6LJ  Website: www.focus4trial.org

MRC CTU AT UCL STAFF

<table>
<thead>
<tr>
<th>Role</th>
<th>Name</th>
<th>Tel:</th>
<th>Email</th>
</tr>
</thead>
<tbody>
<tr>
<td>Trial Managers</td>
<td>Sharandeep Bhogal</td>
<td>020 7670 4805</td>
<td><a href="mailto:d.fisher@ucl.ac.uk">d.fisher@ucl.ac.uk</a></td>
</tr>
<tr>
<td></td>
<td>Gosala Gopalakrishnan</td>
<td>020 7670 4778</td>
<td></td>
</tr>
<tr>
<td>Data Manager</td>
<td>Emma Yates</td>
<td>020 7670 4861</td>
<td></td>
</tr>
<tr>
<td></td>
<td>Bart Przybyl</td>
<td>020 7670 4697</td>
<td></td>
</tr>
<tr>
<td>Project Manager</td>
<td>Anna Bara</td>
<td>020 7670 4643</td>
<td></td>
</tr>
<tr>
<td>Statistician</td>
<td>David Fisher</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Trial Physician</td>
<td>Dr Jenny Seligmann</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Project Lead</td>
<td>Dr Louise Brown</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Programme Lead</td>
<td>Professor Richard Kaplan</td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

For a full list of FOCUS4 contacts and collaborators, please refer to Appendix VI of the FOCUS4 Master Protocol.

RANDOMISATIONS
To randomise, call MRC CTU, Monday to Friday 9 am till 5 pm excluding Bank Holidays
Tel: 020 7670 4777

SAE REPORTING
Within 24 hours of becoming aware of an SAE, please fax a completed SAE CRF to the MRC CTU at UCL on:
Fax: 020 7670 4818

CLINICAL QUERIES
Please direct all queries to the MRC CTU at UCL in the first instance; clinical queries will be passed to the Clinical Trial Fellow or the Chief Investigators via the Trial Managers.
### SUMMARY OF FOCUS4-C

<table>
<thead>
<tr>
<th>SUMMARY INFORMATION TYPE</th>
<th>SUMMARY DETAILS</th>
</tr>
</thead>
<tbody>
<tr>
<td>Acronym</td>
<td>FOCUS4-C</td>
</tr>
<tr>
<td>Note</td>
<td>The FOCUS4 Trial Programme includes separate comparisons for patients from different molecular cohorts. This protocol is for FOCUS4-C and should be used in conjunction with the FOCUS4 Master Protocol.</td>
</tr>
</tbody>
</table>
| Long Title of Trial      | A randomised controlled comparison of AZD1775 (WEE1 inhibitor) versus active monitoring in patients with colorectal cancer that is stable or responding after first-line treatment in two biomarker groups:  
|                          | - Patients with H3K36me3 loss tumours  
|                          | - Patients with tumours mutated at both p53 and RAS |
| Version                  | 5.0             |
| Date                     | 5th April 2018  |
| MRC CTU ID               | CR13            |
| ISRCTN #                 | ISRCTN90061546  |
| EudraCT #                | 2012-005111-12  |
| CTA #                    | 00316/0245/001-0001 |
| REC #                    | 13/SC/0111      |
| Type of Participants to be Studied | Adult patients with inoperable metastatic or locally advanced colorectal cancer (CRC) suitable for intermittent chemotherapy who either have tumours with loss of H3K36me3 or have tumours with both a RAS mutation and a p53 mutation. |
| Trial Design and Interventions to be Compared | FOCUS4 is a prospective, molecularly stratified, adaptive, multi-site programme of randomised controlled comparisons for patients with advanced or metastatic colorectal cancer. This protocol describes the procedures for FOCUS4-C, which is within the FOCUS4 Trial Programme (please refer to FOCUS4 Master Protocol).  
|                          | The FOCUS4 Master Protocol describes the patient identification, registration, biomarker testing and first-line treatment procedures. Following the biomarker panel results, patients are offered entry into a series of comparisons according to their molecular cohort. Each comparison within these patient molecular cohorts will compare an intervention with a standard therapy control. This protocol describes FOCUS4-C which is for patients whose tumour exhibits either:  
|                          | - H3K36me3 loss OR  
<p>|                          | - Mutations of both RAS and p53 |</p>
<table>
<thead>
<tr>
<th></th>
<th></th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Intervention:</strong></td>
<td>AZD1775 (WEE1 inhibitor)</td>
</tr>
<tr>
<td><strong>Control:</strong></td>
<td>Active Monitoring</td>
</tr>
<tr>
<td><strong>Trial Hypothesis</strong></td>
<td>AZD1775 treatment compared with active monitoring shows improved progression free survival in patients with colorectal cancer that have H3K36me3 loss or are RAS mut and p53 mut, which have responded or remained stable after first-line treatment.</td>
</tr>
<tr>
<td><strong>Primary Outcome Measure(s)</strong></td>
<td>Progression Free Survival (PFS) defined as death from any cause or progression of disease according to RECIST v1.1 criteria. An additional primary outcome of overall survival (OS) may be evaluated.</td>
</tr>
<tr>
<td><strong>Secondary Outcome Measure(s)</strong></td>
<td>Safety, toxicity and tumour response.</td>
</tr>
<tr>
<td><strong>Randomisation</strong></td>
<td>Randomisation will use the method of minimisation including a random element and stratified on a list of potentially prognostic factors (see Master Protocol). The randomisation allocation ratio will be 2:1 (AZD1775: Active Monitoring).</td>
</tr>
</tbody>
</table>
| **Number of Participants to be Studied** | H3K36me3 loss cohort:  Stage I – 34  Stage II – 55  
|                                     | RAS and p53 mutation cohort:  Stage I – 70  Stage II – 113 |
| **Duration**      | 28 months                                                       |
| **Ancillary Studies/Sub-studies** | 1) Assessment of further markers of DNA repair deficiency and/or sensitivity to WEE1 inhibitors.  
|                                     | 2) Identification of acquired resistance to therapy through circulating free tumour DNA.  
|                                     | 3) Assessment of pre- and post-treatment metastatic biopsy material for resistance mechanism evaluation.  
|                                     | Please refer to the FOCUS4 Master Protocol for Ancillary Studies applicable for the FOCUS4 Trial Programme. |
| **Sponsor**       | Medical Research Council                                        |
| **Funder**        | FOCUS4 is funded by the MRC/NIHR EME programme and CRUK. Funding for drug and distribution is provided by AstraZeneca in the UK. |
| **Chief Investigators** | **FOCUS4 Trial Programme:**  Professor Tim Maughan and Professor Richard Wilson.  
|                                     | **FOCUS4-C:**  Professor Tim Maughan, Professor Matt Seymour and Dr Jenny Seligmann. |
Figure 1: Current FOCUS4 Trial Schema 2018 (unshaded cohorts open, shaded cohorts in set-up or development)
Figure 2: FOCUS4-C Schema

Registration and first-line treatment plus interim and end of first-line treatment CT scans
(refer to FOCUS4 Master Protocol for procedure prior to randomisation)

1. Responding or Stable Disease after first-line treatment
2. Tumour H3K36me3 loss or combined RAS and P53 mutations
3. Eligible for FOCUS4-C

Consent for randomisation

Randomisation
Please ensure there is a 3 week washout period between first-line treatment and trial treatment

H3K36me3 loss
- Arm C1: Active Monitoring

RAS & P53 mutations
- Arm C2: AZD1775

Clinic and trial assessments every 3 weeks for safety and toxicity, and every 8 weeks for CT scans and RECIST assessments.

Stop treatment due to progression, cumulative toxicity or patient choice

Treatment after stopping Trial Therapy:
After completion of trial therapy, patients restart first-line treatment at clinical discretion. Patients may be eligible for treatment with oxaliplatin or irinotecan or entry into another clinical trial.
# FOCUS4-C ASSESSMENTS SCHEDULE

## Table 1: FOCUS4-C Assessments Schedule
(The table is also provided in Section 6 - Trial Assessment and Follow-up)

<table>
<thead>
<tr>
<th></th>
<th>Pre-randomisation</th>
<th>Every 3-4 weeks until end of trial treatment *</th>
<th>Every 7-9 weeks until progression</th>
<th>If any SAE occurs</th>
<th>Within 4 weeks of end of trial treatment (progression, toxicity or patient choice)</th>
<th>Follow-up after progression at 3 months and then 6 monthly</th>
</tr>
</thead>
<tbody>
<tr>
<td>Clinical evaluation (including Blood pressure, Pulse rate, weight measurement)</td>
<td>X^</td>
<td>X</td>
<td>X</td>
<td>X</td>
<td>X</td>
<td>X</td>
</tr>
<tr>
<td>WHO PS</td>
<td>X^</td>
<td>X</td>
<td>X</td>
<td>X</td>
<td>X</td>
<td>X</td>
</tr>
<tr>
<td>Pregnancy Test (for women of child bearing potential)~</td>
<td>X^</td>
<td>X</td>
<td>X</td>
<td>X</td>
<td>X</td>
<td>X</td>
</tr>
<tr>
<td>Toxicity (NCI CTCAE)</td>
<td>X^</td>
<td>X</td>
<td>X</td>
<td>X</td>
<td>X</td>
<td>X</td>
</tr>
<tr>
<td>Review use of concomitant medications</td>
<td>X^</td>
<td>X</td>
<td>X</td>
<td>X</td>
<td>X</td>
<td>X</td>
</tr>
<tr>
<td>FBC, U&amp;Es, LFTs, ALT/AST, ALKP</td>
<td>X^</td>
<td>X</td>
<td>X</td>
<td>X</td>
<td>X</td>
<td>X</td>
</tr>
<tr>
<td>CEA, LDH</td>
<td>X^</td>
<td>X</td>
<td>X</td>
<td>X</td>
<td>X</td>
<td>X</td>
</tr>
<tr>
<td>GFR</td>
<td>X^†</td>
<td>X</td>
<td>X†</td>
<td></td>
<td>X</td>
<td>X</td>
</tr>
<tr>
<td>CT Scan</td>
<td>X¶</td>
<td></td>
<td>X</td>
<td></td>
<td>X</td>
<td>X</td>
</tr>
<tr>
<td>RECIST v1.1 response from CT scan</td>
<td>X¶</td>
<td></td>
<td>X</td>
<td></td>
<td>X</td>
<td>X</td>
</tr>
<tr>
<td>ECG</td>
<td>X^</td>
<td>X</td>
<td>X†</td>
<td></td>
<td>X</td>
<td>X α</td>
</tr>
<tr>
<td>Dispensing AZD1775, Diary Card assessment</td>
<td>X=&lt;#</td>
<td></td>
<td></td>
<td></td>
<td>X</td>
<td>X</td>
</tr>
<tr>
<td>CRFs/eCRFs required</td>
<td></td>
<td>Randomisation CRF</td>
<td>Treatment eCRF &amp; Toxicity &amp; Symptom eCRF</td>
<td>Progress eCRF &amp;</td>
<td>SAE Report CRF</td>
<td>Toxicity &amp; Symptom eCRF</td>
</tr>
<tr>
<td>Optional assessments (if specific consent given)</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Circulating free DNA</td>
<td>X‡</td>
<td>X‡</td>
<td>X‡ for progression only</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Optional Tumour biopsy</td>
<td>X§</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td>X§</td>
</tr>
</tbody>
</table>

### Key for symbols

- ^ Within 14 days prior to randomisation
- † Not required if calculated creatinine clearance >50ml/min (using local protocols)
- ¶ Pre-randomisation CT (baseline comparator scan) within 28 days prior to randomisation
For patients on Arm C2 (AZD1775) only. ECG to be performed within 1 week prior to starting each cycle while on AZD1775 but not required prior to cycle 1 as pre-randomisation ECG is available.

Women of child bearing potential on Arm C2 (AZD1775) must have a negative pregnancy test 7 days prior to randomisation and every 3 week while on trial treatment. They must also be using adequate contraception for two weeks prior to starting trial treatment, while receiving AZD1775 and for one month following end of trial treatment. Male patients should be willing to abstain or use barrier contraception (i.e., condoms) for the duration of the trial treatment and for 3 months after stopping AZD1775.

Patients in the active monitoring arm still require appointments with the Research Nurse every 4 weeks to check they are well.

If clinically indicated by the event

Until patient stops trial treatment due to progression, cumulative toxicity or patient choice

Optional blood samples not required if patient withholds consent for molecular research

Optional tumour biopsies not required if patient withholds consent to additional biopsies. If patient consents to this sub-study, biopsy should be taken after randomisation (prior to starting trial treatment) and on progression.
# CONTENTS

GENERAL INFORMATION .................................................................................................. II
FOCUS4-C ADMINISTRATION .................................................................................... III
SUMMARY OF FOCUS4-C .......................................................................................... IV
FOCUS4 TRIAL SCHEMA ........................................................................................... VI
FOCUS4-C ASSESSMENTS SCHEDULE ..................................................................... VIII
CONTENTS .................................................................................................................... X
1. BACKGROUND ......................................................................................................... 1
   1.1 FOCUS4-C ........................................................................................................... 1
   1.2 INTRODUCTION ................................................................................................. 1
   1.3 CHOICE OF THERAPEUTIC INTERVENTION: .................................................... 3
       1.3.1 RATIONALE .............................................................................................. 3
       1.3.2 AZD1775 PHARMACOKINETICS AND PHARMACODYNAMICS .............. 6
2. SELECTION OF SITES/CLINICIANS ..................................................................... 9
   2.1 CATEGORISATION OF SITES .......................................................................... 9
   2.2 SITE INCLUSION CRITERIA ............................................................................. 9
   2.3 SITE EXCLUSION CRITERIA ........................................................................... 9
   2.4 APPROVAL AND ACTIVATION ....................................................................... 9
       2.4.1 FOCUS4 TRIAL PROGRAMME APPROVALS AND ACTIVATION .......... 9
       2.4.2 FOCUS4-C REQUIRED SITE TRAINING .............................................. 9
       2.4.3 FOCUS4-C APPROVALS AND ACTIVATION ........................................ 10
3. SELECTION OF PATIENTS FOR FOCUS4-C ......................................................... 11
   3.1 REGISTRATION ................................................................................................ 11
   3.2 PATIENT SCREENING AND INFORMATION-GIVING PROCESS ...................... 11
   3.3 PATIENT INCLUSION AND EXCLUSION CRITERIA FOR RANDOMISATION ...... 12
       3.3.1 INCLUSION CRITERIA ............................................................................ 12
       3.3.2 EXCLUSION CRITERIA ........................................................................... 13
   3.4 ACTIONS PRIOR TO RANDOMISATION .............................................................. 14
4. SELECTION OF PATIENTS FOR FOCUS4-C RANDOMISATION ............................. 16
   4.1 RANDOMISATION ............................................................................................ 16
   4.2 RANDOMISATION CODES .............................................................................. 16
   4.3 TRANSLATIONAL RESEARCH SAMPLE COLLECTION .................................. 16
   4.4 CO-ENROLMENT GUIDELINES ..................................................................... 17
   4.5 REGISTERED PATIENTS WHO DO NOT CONSENT TO RANDOMISATION ...... 17
5. TREATMENT OF PATIENTS IN FOCUS4-C .......................................................... 18
   5.1 INTRODUCTION AND TREATMENT ALLOCATION ....................................... 18
   5.2 TREATMENT AND CT SCAN SCHEDULE ......................................................... 18
       5.2.1 ACTIVE MONITORING ARM ................................................................ 18
       5.2.2 AZD1775 ARM ................................................................................. 18
       5.2.3 TOLERABILITY PHASE AND PLANNED REVIEW OF AZD1775 STARTING DOSE 20
5.2.4 DOSE MODIFICATIONS, INTERRUPTIONS & DISCONTINUATIONS FOR THE AZD1775 ARM ........................................ 21
5.3 PROTOCOL TREATMENT DISCONTINUATION ................................................................. 21
5.4 COMPLIANCE & ADHERENCE .................................................................................. 22
5.5 TREATMENT DATA COLLECTION ........................................................................ 22
5.6 TREATMENT ON PROGRESSION ........................................................................ 22
5.7 AZD1775 ................................................................................................................. 23
5.7.1 PRODUCTS ........................................................................................................ 23
5.7.2 DISPENSING AND DRUG SUPPLIES .................................................................. 23
5.7.3 ACCOUNTABILITY & UNUSED DRUGS ................................................................ 24
5.8 OVERDOSE OF TRIAL MEDICATION ...................................................................... 24
5.9 MEDICATIONS OUTSIDE OF THE TRIAL ................................................................. 24
5.9.1 PERMITTED AND PROHIBITED CONCOMITANT MEDICATIONS .................. 25
5.9.2 MEDICATIONS AND FOOD TO BE USED WITH CAUTION ............................ 25
6 TRIAL ASSESSMENTS & FOLLOW-UP FOR FOCUS4-C ........................................... 27
6.1 CASE REPORT FORM (CRF) RETURN GUIDELINES .................................................. 28
6.2 PROCEDURES FOR ASSESSING EFFICACY ............................................................. 28
6.3 PROCEDURES FOR ASSESSING SAFETY .................................................................. 29
6.4 PROCEDURES FOR ASSESSING QUALITY OF LIFE .............................................. 29
6.5 OTHER ASSESSMENTS ............................................................................................ 29
6.6 EARLY STOPPING OF FOLLOW-UP ......................................................................... 29
6.7 PATIENT REFERRALS AND TRANSfers .................................................................. 30
6.8 LOSS TO FOLLOW-UP .............................................................................................. 30
7 SAFETY REPORTING .................................................................................................. 31
7.1 DEFINITIONS ......................................................................................................... 31
7.1.1 MEDICINAL PRODUCTS .................................................................................... 31
7.1.2 ADVERSE EVENTS ............................................................................................. 31
7.1.3 EXEMPTED ADVERSE EVENTS ....................................................................... 31
7.1.4 DISEASE-RELATED EVENTS ............................................................................. 33
7.2 OTHER NOTABLE EVENTS ...................................................................................... 33
7.2.1 PREGNANCY ..................................................................................................... 33
7.3 INVESTIGATOR RESPONSIBILITIES ....................................................................... 33
7.3.1 INVESTIGATOR ASSESSMENT ........................................................................ 34
7.3.2 NOTIFICATION PROCEDURE ......................................................................... 36
7.4 MRC CTU AT UCL RESPONSIBILITIES ................................................................ 36
8 QUALITY ASSURANCE & CONTROL ...................................................................... 38
8.1 RISK ASSESSMENT ................................................................................................. 38
8.2 CENTRAL MONITORING AT MRC CTU AT UCL .................................................. 38
8.3 ON-SITE MONITORING ........................................................................................ 38
8.3.1 DIRECT ACCESS TO PATIENT RECORDS ...................................................... 38
8.3.2 CONFIDENTIALITY ............................................................................................ 38
9 STATISTICAL CONSIDERATIONS ........................................................................... 39
9.1 METHOD OF RANDOMISATION ............................................................................. 39
9.2 OUTCOME MEASURES ........................................................................................... 39
9.3 SAMPLE SIZE ........................................................................................................ 39
9.4 INTERIM MONITORING & ANALYSES ................................................................. 39
9.5 ANALYSIS PLAN (BRIEF) ..................................................................................... 40
10 ANCILLARY STUDIES ........................................................................................................42
11 OTHER ISSUES .............................................................................................................43
12 PROTOCOL AMENDMENTS .........................................................................................44
13 REFERENCES ...............................................................................................................46
1. BACKGROUND

1.1 FOCUS4-C

FOCUS4-C is for patients from the ‘synthetic lethality’ molecular cohort (those who demonstrate either H3K36me3 loss, or RAS mutation and p53 mutation). This comparison protocol therefore covers the processes following identification of patients into this molecular cohort and their eligibility for FOCUS4-C. No other molecular cohort or subsequent comparison will be referred to in this protocol. The FOCUS4 Master Protocol documents the generic aspects of the FOCUS4 Trial Programme and processes leading up to entry into FOCUS4-C including registration into FOCUS4 and biomarker stratification and should be referred to for these pre-randomisation processes.

1.2 INTRODUCTION

Genome instability is considered an ‘enabling characteristic’ of cancer[1] and a wide range of DNA damage response and repair genes are mutated in cancer cells[2]. Cells have evolved strict surveillance mechanisms that detect DNA damage, coordinate cell cycle progression and coordinate DNA repair pathways to ensure genome integrity prior to replication. Normally, only cells with stable genomes will proceed to mitosis, thus avoiding the generation of genetically aberrant progeny cells; detection of abnormality leads to stalling of the cell cycle and initiation of DNA repair or, if damage is extensive or DNA repair efforts fail, then induction of apoptosis.

The response to DNA damage is mediated through complex pathways, involving sensors, signal transducers and effectors which trigger the mobilisation of cell-cycle checkpoints and DNA repair proteins. Cell-cycle checkpoints are mechanisms to halt cell cycle progression and allow more time for repair of damaged DNA prior to critical phases of replication or entry into mitosis. Defects in cell-cycle checkpoint control are seen in certain hereditary cancer syndromes and at early stages of malignant transformation.

ATM and ATR kinases are key responders to DNA replication forks which have stalled due to DNA damage and replication stress. They are activated through phosphorylation, and in their turn activate downstream effectors including Chk1/2 kinases and p53. These then delay DNA replication, prevent entry into mitosis, and promote DNA repair (Figure 1.1). The mechanisms involved in these processes include cell cycle checkpoint control at G1/S, S and G2/M, direct regulation of DNA replication, and DNA repair through the homologous recombination repair and cross-link repair pathways.

Cell cycle regulators have been identified as promising therapeutic targets in cancer in two contexts:

- In combination with chemotherapy or radiotherapy, where inhibiting cell cycle regulation may drive damaged cells inappropriately through the cell cycle without repair.
- As monotherapy in cancers with defective DNA repair mechanisms, where we might anticipate a ‘synthetic lethality’ interaction.
Figure 1.1 The DNA damage checkpoints. In response to DNA damage or replication fork stalling, two signalling cascades mediated by ATM and ATR arrest the cells in G1/S, S or G2/M by modulating the activity of cyclin-dependent kinases (CDK1 and CDK2)

‘Synthetic lethality’ as a therapeutic approach

Synthetic lethality is a generic term for when two pathways together perform an essential function, and the loss of one pathway (due to mutation or pharmacological inhibition) is tolerated, but loss of both pathways leads to cell death. For example, cells with a non-functioning G1/S checkpoint are viable, provided they have retained function at the intra-S-phase or G2/M checkpoint; however, loss of both causes cell death.

Synthetic lethality may be exploited to selectively target tumour cells where one key function is already absent due to genetic alteration[3]. The most clinically advanced example of this is with PARP inhibitors in BRCA-deficient cancers. BRCA proteins are required for DNA repair by homologous recombination (HR); PARP1 is a key component of DNA base-excision repair. Cells can tolerate loss of one of these pathways; however, in tumours lacking HR due to loss of BRCA, treatment with a PARP inhibitor leads to high levels of unrepaired dsDNA breaks during S-phase and subsequent cell death[4, 5]. BRCA1 or 2 deficient cells were 1000-fold more sensitive to PARP inhibition than wild-type cells in vitro[5]; subsequent clinical studies have demonstrated an enhanced effect of PARP inhibition in patients with BRCA1/2 deficient breast[6] or ovarian cancers[7-9]. And in a phase II trial of olaparib monotherapy in metastatic prostate cancer, 14/16 (88%) patients with tumours bearing known DNA repair gene aberrations responded, compared with only 2/16 (6%) patients without those aberrations [10].
1.3 CHOICE OF THERAPEUTIC INTERVENTION:

In FOCUS4-C, the novel WEE1 inhibitor AZD1775 will be compared to Active Monitoring.

WEE1 is a nuclear tyrosine kinase which delays entry into mitosis by inhibiting activity of the cyclin-dependent kinases CDK1 and CDK2 through tyrosine 15 phosphorylation. CDK inactivation can lead to cell cycle arrest at G1/S and G2/M (Figure 1.1) \[11, 12\]. In this way, WEE1 activation helps prevent entry into mitosis until DNA replication is complete and histone synthesis is complete (H2B y37-phosphorylation): these actions maintain optimal DNA-histone stoichiometry prior to mitotic entry \[11, 13\]. WEE1 may also affect the intra-S-phase checkpoint by modulating CDK1/2 at replication origins to block replication initiation \[14\]. The central role of WEE1 in integrating various aspects of cell cycle progression, histone synthesis and genomic stability makes it an attractive therapeutic target.

Inhibition of WEE1 has been shown to cause unscheduled entry into mitosis, and aberrant firing of replication origins leading to dNTP shortage and replication stress \[11\]. Additionally, WEE1 deficient cells accumulate DNA damage during S-phase due to interference with DNA-damage repair mechanisms.

AZD1775 is the first small molecule inhibitor of WEE1 kinase. Clinical studies early in the development of AZD1775 (previously MK1775) focused on enhancing the activity of chemotherapy, based upon the rationale that abrogation of cell cycle checkpoints would push damaged cells into premature mitosis with unrepaired DNA, with resultant cell death. In cell models AZD1775 enhanced the anti-tumour activity of various chemotherapeutic agents including 5-FU and gemcitabine \[15-17\]. However, there is also interest in its use as a single agent to generate synthetic lethality in tumours with DNA repair defects.

1.3.1 RATIONALE

1. Testing of AZD1775 in the H3K36me3 loss biomarker group

**Hypothesis:** AZD1775 will induce synthetic lethality in tumours with H3K36me3 loss, with reduction in RRM2 levels, critical depletion of dNTP pools and subsequent replication stress during S-phase. Patients with H3K36me3-loss tumours will therefore have improved PFS with AZD1775 maintenance therapy compared with Active Monitoring.

SETD2 (Chr. 3p) is frequently mutated in cancer \[30\] and is observed in multiple cancer sites \[31, 32\]. SETD2 encodes histone H3K36me3 methyltransferase, generating histone H3K36 trimethylation (H3K36me3), whose functions include DNA repair, and stem cell regulation. Loss of H3K36me3 can be caused by at least 3 different mechanisms: loss of SETD2 function; overexpression of oncogene KDM4A (the histone H3K36me3 demethylase), or mutations in histone H3.3. These alterations have been described in several cancer types and are associated with poor prognosis.
In fission yeast cells, a synthetic interaction was observed between loss of Set2 (the human SETD2 homologue) and WEE1 (the human WEE1 homologue) [33]. SETD2-deficient cells were hypersensitive to treatment with AZD1775; restoring SETD2 led to increased H3K36me3 levels and reduced sensitivity. SETD2 deficient cells treated with AZD1775 showed disturbance in S-phase, with cells accumulating as non-replicating S-phase cells, but did not show evidence of premature mitosis [33].

Treating H3K36me3-loss tumours with AZD1775 leads to reduction in ribonuclease reductase (RNR) transcription and RRM2 protein levels. This synthetically lethal interaction results because two pathways regulate RRM2 levels (Figure 1.2): H3K36me3 transcriptionally activates RRM2; while WEE1 suppresses CDK-dependent degradation of RRM2 during DNA replication. Simultaneous inhibition of both pathways leads to a dramatic reduction in RRM2, pushing dNTP levels to critically low levels. Cells subsequently undergo replication fork arrest, leading to DNA damage at stalled forks (through MUS81 endonuclease), which triggers cell death [33].

![Figure 1.2: Demonstrating both pathways regulating RRM2, the target of the synthetically lethal interaction between H3K36me3 loss and WEE1 inhibition](image)

Importantly this mechanism is independent of p53 status: it is seen in p53 wild-type cell lines, and SETD2 deficiency did not affect p53 activity. BRCA deficient cells were not hypersensitive to WEE1 inhibition, suggesting that WEE1’s role in HR is not greatly impacting the synthetic interaction in this setting [33].

As a clinically relevant synthetic interaction between H3K36me3 loss and WEE1 inhibition has been demonstrated it was important to develop a clinically usable single biomarker that could identify all H3K36me3-deficient tumours. A monoclonal antibody for H3K36me3 has been developed which is able to distinguish SETD2 deficient tumours from proficient tumours. This antibody is also worked up on FFPE tumour microarrays and will be used in this study to select patients for this comparison.
2. Testing AZD1775 in the RAS mut/p53 mut biomarker group

Hypothesis: **AZD1775 will induce synthetic lethality in RAS/p53 mutant tumours as they demonstrate replication stress and defective G1 checkpoints. Patients in this biomarker group will have improved PFS with maintenance AZD1775 compared with Active Monitoring.**

**P53** is a key regulator of the G1 checkpoint and is one of the most frequently mutated genes in cancer [18]. Loss of function of p53 leads to abrogation of the G1 checkpoint, and dependence on the intra-S and G2/M checkpoints to detect damage and initiate repair. p53-deficient cells are therefore more susceptible to treatment targeting the G2 checkpoint [19]. In preclinical studies AZD1775 possessed preferential killing effect in p53-deficient compared with p53-wild type tumours [16, 17].

A recently reported phase 2 study compared AZD1775 in combination with carboplatin in p53-mutant ovarian cancer resistant to first-line therapy [20]. 6/22 (27%) evaluable patients had a PR and a further 9 (41%) had SD. Several studies investigating AZD1775 in combination with chemotherapy are now ongoing.

**RAS** is mutated in around 45% patients with advanced colorectal cancer (aCRC). It has been an elusive therapeutic target, made difficult due to its biochemistry and structure and highly complex nature of the signalling cascades, and novel treatment strategies are urgently required. RAS has well recognised actions through downstream MAPK-AKT pathway signalling; however, it is also able to drive cell cycle progression, leading to replication stress during S-phase. In preclinical studies, mutant RAS drives cells into S-phase through regulation of the CDK4 or CDK6 complex, and provides sustained mitogenic signals through sustained CDK2 activity. These effects activate the replication stress response including checkpoint activation [21]. Mutant RAS can also stimulate ATR with subsequent activation of downstream effectors including Chk1/2 kinases and p53 [22, 23]. Thus, mutated RAS can lead to replication stress and is dependent upon functional checkpoints.

For all these reasons there is a strong rationale to expect synthetically lethal interactions of mutated RAS with genes regulating mitotic functions and cell cycle checkpoints [24-26]. AZD1775 has been identified as a potentially useful drug in RAS-mutant cancers: WEE1 plus mTOR inhibition in KRAS-mutant colorectal cancer was associated with inhibition of cell growth in *in vitro* models [27].

Theoretically a tumour with both p53 loss and RAS mutant will be highly vulnerable to AZD1775 inhibition: such a tumour will have G1 checkpoint failure and evidence of replication stress and will be reliant on the intra-S-phase and G2/M checkpoints to allow DNA repair. In a post-AZD1775 treatment biopsy of a p53 mut/RAS-mut patient, the tumour had invoked both mechanisms of response to WEE1 inhibition with evidence of DNA damage response with Cdk2 activation and mitogenic signalling, compared with baseline [28]. In lung cancer models RAS activation and loss of p53 function appeared to work collaboratively to cause dysregulation of cell cycle progression and
inhibition of apoptosis, potentially through activation of NFkB [29]. Hence the combination of these 2 mutations are likely to produce a more potent synthetically lethal interaction with AZD1775 than either alone.

1.3.2 AZD1775 PHARMACOKINETICS AND PHARMACODYNAMICS

AZD1775 is an orally active, highly selective, potent (IC_{50} = 5.18 nM) ATP competitive, small-molecule inhibitor of the WEE1 kinase. In cell line studies (including p53 wild-type colorectal adenocarcinoma) AZD1775 monotherapy showed strong induction of the DNA damage response [34]. In a p53 deficient colorectal cancer cell line, AZD1775 in combination with gemcitabine inhibited CDC2-Tyr15 phosphorylation (the direct substrate of WEE1) with an EC value of 84.87 nM(+/-17.04), demonstrating inhibition of WEE1 cellular activity. Therefore single-agent preclinical activity has been demonstrated, independent of p53 status, suggesting that other mechanisms may contribute to AZD1775 sensitivity [34, 35].

In vivo models demonstrate oral bioavailability and anti-tumour effects of AZD1775. In nude mouse xenograft models (non-small cell lung cancer), daily treatment with AZD1775 monotherapy led to 51% tumour regression. In pharmacokinetic and dynamic evaluation in xenograft tumour tissue, continuous treatment of AZD1775 caused a dose-dependent reduction in phospho-CDC2-Tyr15.

Following single oral administration in humans, the peak plasma drug concentration (C_{max}) is observed after 3-4 hours. Post-peak plasma concentrations decline approximately mono-exponentially with T_{1/2} in the region of 10 hours. Exposure as measured by C_{max} and AUC_{0-∞} increased in a dose-proportional manner over the dose range of 325 to 1300mg.

Preliminary investigation of drug-drug interactions (DDIs) in Study PN001 suggest a 40-60% increase in the exposure of AZD1775 in the presence of aprepitant (moderate CYP3A4 inhibitor), but no effect with the concomitant administration of steroids (moderate CYP3A4 inducers). Preliminary studies also suggested that the pre-marketed Oral Formulation of AZD1775 was similar to that of the Fit-For-Purpose formulation. Based on the preliminary comparison of the results of AZD1775 PK parameters at the 225 mg dose, PK estimates in Asian patients were somewhat higher than in Western patients, with first-dose C_{max} and AUC values 45% and 35% higher, respectively, in the Asian than the Western population (Study PN011). At steady state, a similar trend toward higher exposure in Asian patients was observed. Additional analysis/investigation will be conducted based on the emerging data to understand the exposure differences between the populations.

In an ongoing NCI cohort study (NCT01748825), once-daily (O.D.) doses of 200-300 mg have been evaluated without reaching dose-limiting toxicity. Preliminary PK data from O.D. dosing shows non-linearity, with exposure (C_{max} and AUC) increasing proportionally more than dose, between 200 and 300 mg, after both the first and subsequent doses. Comparison with twice-daily (B.I.D.) dosing in Study NCT02482311 shows that AUC_{0-24} on day 5 at 200-250 mg O.D. dose is similar to that achieved with B.I.D. dosing at 175 mg; however, at 300 mg O.D., the AUC_{0-24} on day 5 exceeds that observed with B.I.D. dosing at 200 mg (22,498 versus 19,632 nM.h). (AstraZeneca data on file).
As of 11 November 2016, approximately 551 patients had been exposed to AZD1775 in AstraZeneca or Merck-sponsored clinical studies, with a further 350 patients having received AZD1775 as part of academically-sponsored research. Of the 551 patients, 103 received AZD1775 monotherapy, 407 patients received AZD1775 in combination with cytotoxic chemotherapy agents and the remaining 41 patients received AZD1775 in combination with targeted therapies MEDI4736 or olaparib. These patients have received single doses per cycle as high as 1300 mg of AZD1775 as monotherapy, 325 mg of AZD1775 in a single-dose in combination with chemotherapy, and 325 mg B.I.D. in a multiple-dose regimen in combination with chemotherapy. Please refer to the current version of the AZD1775 Investigator’s Brochure.

The completed or terminated early studies include:

**NCT 00648648:** a first-time-in-patients (FTIP), Phase I, dose-escalation study evaluating AZD1775 both as monotherapy and combination therapy with gemcitabine, cisplatin, or carboplatin in adult patients with advanced solid tumours (Part 1 & 2 only completed)

**NCT 010477007:** a Phase I, dose-escalation study evaluating AZD1775 as monotherapy (Part 1), combination therapy with 5-FU (Part 2), and combination therapy with 5FU plus cisplatin (Part 3) in adult Japanese patients with advanced solid tumours was terminated early due to portfolio prioritisation in oncology at Merck after 3 patients had been enrolled in Part 1 and 8 patients had been enrolled in Part 2. Part 3 was not initiated.

**NCT 01076400:** a Phase I/IIa, dose-escalation study evaluating AZD1775 in combination with topotecan plus cisplatin in adult patients with cervical cancer was terminated early due to portfolio prioritisation in oncology at Merck after 7 patients had been enrolled in the dose-escalation part of the study. The Phase IIa part was not initiated.

**NCT 01748825:** a Phase I study of single-agent AZD1775, in patients with refractory solid tumours, sponsored by the National Cancer Institute (NCI) Cancer Therapy Evaluation Program in collaboration with AstraZeneca and Merck. This study reported AZD1775 monotherapy activity in patients carrying BRCA mutations for the first time. An ongoing O.D. dosing of schedule component of this study is still ongoing.

**NCT 02087176:** a lead-in Phase II multicentre, randomised, double-blind study comparing AZD1775 plus docetaxel with placebo plus docetaxel in previously treated patients with non-small-cell lung cancer (NSCLC).

**NCT 02087241:** a Phase II study of AZD1775 plus pemetrexed and carboplatin followed by a randomised comparison of pemetrexed and carboplatin with or without AZD1775 in patients with previously untreated stage IV non-squamous NSCLC.

In study **NCT 006486648**, of 176 evaluable patients who received AZD1775 (either single or multiple doses) as monotherapy or in combination with gemcitabine, cisplatin, or carboplatin, a partial response (PR) (confirmed and unconfirmed) was observed in 17 (9.7%) patients, and stable disease (SD) was observed in 94 (53.4%) patients (AZD1775 Investigator’s Brochure [IB]).
Nine patients received AZD1775 monotherapy. Single ascending doses of AZD1775 up to 1300 mg were well tolerated; the maximum tolerated dose (MTD) was not established.

In study **NCT 010477007**, patients in Part 1 received single-cycle B.I.D. dosing of AZD1775 for 5 days at 1 of 2 dose levels as monotherapy. A cohort of 3 patients was enrolled at the starting dose level of AZD1775 65 mg B.I.D. and no serious adverse events (SAEs) were experienced.

In study **NCT 01748825**, patients received single-agent AZD1775 B.I.D. over 2½ days per week for 2 weeks in 3-week treatment cycles. Twenty-five patients were enrolled to determine the MTD using a 3+3 design. The MTD was established at 225 mg by mouth (PO) B.I.D for 5 doses on Weeks 1 and 2 of a 3-week schedule. Six patients with BRCA-mutated solid tumours were enrolled at the MTD. Partial responses were confirmed in 2 of the patients carrying BRCA mutations (ovarian cancer patient and head/neck cancer patient). Paired tumour biopsies were obtained from 5 patients treated at the MTD at baseline and after the 5th AZD1775 dose to determine the levels of pY15-Cdk and γH2AX. The biopsies showed a decrease in pY15-Cdk levels (2/5 paired biopsies). The same biopsies were analysed for increases in γH2AX, an indicator of DNA damage. Three of the 5 biopsy pairs showed an increase in γH2AX levels. DNA damage response was observed in this study through provided paired tumour biopsies (Do *et al* 2015). Additional information may be found in the current version of the AZD1775 IB.
2 SELECTION OF SITES/CLINICIANS

2.1 CATEGORISATION OF SITES

In FOCUS4, due to the early stage of development of the agents being tested, sites will be assessed and categorised based on their facilities and experience in phase 1 and 2 clinical trials. For details on the definitions of the categories, criteria and their assessment, please refer to the FOCUS4 Master Protocol.

FOCUS4-C will utilise the novel WEE1 inhibitor AZD1775. Given the current safety data on this compound, patients in FOCUS4-C can only be consented and treated at sites that are assessed to be:
- Level 3
- Level 2 (after an initial tolerability phase)

The trial includes an initial phase to assess tolerability in 12-18 patients, during which only Level 3 sites may randomise and treat patients in FOCUS4-C. It is anticipated that the comparison will be extended to Level 2 sites providing tolerability is confirmed. Sites which are not treating patients in FOCUS4-C (Level 1 sites, and Level 2 sites during the initial phase) are asked to refer patients to an open Level 3 site for FOCUS4-C randomisation consent, randomisation and treatment. Patients can be referred back to the original site when deemed safe for the patient at the discretion of the randomising site clinician. If patients do not wish to travel to other sites from the registering site or do not want to enter FOCUS4-C, these patients may be offered entry into FOCUS4-N (Non-stratified).

For further details on this comparison, please refer to the FOCUS4-N protocol.

Later in the trial, it may be assessed that FOCUS4-C may be able to open in Level 1 sites. The MRC CTU at UCL will inform all sites if this occurs. For the current situation please refer to the FOCUS4 website, www.focus4trial.org

2.2 SITE INCLUSION CRITERIA

Please refer to Section 2.2 of the Master Protocol for a full list of Level 2 and 3 inclusion criteria.

2.3 SITE EXCLUSION CRITERIA

- Site assessed to be a Level 1 site or Level 2 during the initial tolerability phase
- Lack of any of the site inclusion criteria defined in section 2.2 of the Master Protocol

2.4 APPROVAL AND ACTIVATION

2.4.1 FOCUS4 TRIAL PROGRAMME APPROVALS AND ACTIVATION

Please refer to the FOCUS4 Master Protocol for documentation required for site activation.

2.4.2 FOCUS4-C REQUIRED SITE TRAINING

To participate in FOCUS4-C, sites must have participated in site training with the attendance of at least the site PI, one research nurse and a pharmacy representative. This may be incorporated
through attendance of the FOCUS4 investigator/Launch meetings, a site initiation visit or participation in a site training teleconference. It will be the responsibility of those who attend the training to disseminate the training to other site personnel.

For sites already open, further training on FOCUS4-C will be provided.

2.4.3 FOCUS4-C APPROVALS AND ACTIVATION

For new sites, following receipt of the documents listed in Section 2.5.1 of the Master Protocol at the MRC CTU at UCL and all required training being performed; written confirmation of FOCUS4 activation will be sent to the PI and all relevant site personnel. This will confirm the site’s assessment of level and therefore if they are activated to participate in FOCUS4-C. An accreditation pack will be provided to the site containing documents for FOCUS4-C once when they are activated.

The site pharmacist will also be informed of the site’s activation to FOCUS4-C and be sent the pharmacy packs containing required relevant information for drug dispensing.

For sites already open when FOCUS4-C is activated, this comparison will be activated as a substantial amendment to the FOCUS4 Trial Programme. Following this substantial amendment (and any others that occur), sites will be notified of relevant documents and training required and if and when they are able to participate. Further accreditation packs may be circulated as a result to update trial documentation.
3 SELECTION OF PATIENTS FOR FOCUS4-C

3.1 REGISTRATION

The process of registration and biomarker panel testing are documented in the FOCUS4 Master Protocol. This FOCUS4-C protocol documents the process following biomarker testing and identification of patients as eligible for FOCUS4-C. An End of Registration eCRF in the registration MACRO database must be completed for all patients who are registered into FOCUS4 regardless of whether they subsequently enter one of the randomised comparisons.

3.2 PATIENT SCREENING AND INFORMATION-GIVING PROCESS

The process for giving information to patients about FOCUS4 has a stepped approach to reduce the possibility of information overload. A two-step process has been designed to overcome this. Patient Information Sheet (PIS) 1 will be given to the patient at registration and PIS2 will be given when the biomarker panel results are known and the interim CT scan during first-line treatment (preferably performed 8 weeks after start of first-line treatment) has shown stable or responding disease. PIS2 should be given at the Level 1 site (or Level 2 site during the tolerability phase) prior to referral to Level 2 or 3 sites. This will allow patients as much time as possible to consider participation in any comparison offered to them subsequently. Therefore, PIS2 is specific to each individual comparison. Please refer to Figures 3.1 and 3.2 of the Master Protocol for a description of when the consent process should be initiated at Level 1, 2 or 3 sites.

PIS2 for FOCUS4-C (PIS2-C) describes in detail the trial randomisation that is specific to patients with known H3K36me3 loss or combined RAS and p53 mutations. This will provide details of the potential advantages and disadvantages of being allocated to either control (Active Monitoring) or treatment (AZD1775) arm.

PIS2-C can only be provided:

- When the biomarker panel results are complete and received from the MRC CTU at UCL.
- MRC CTU at UCL confirm that the patient is in the synthetic lethality cohort and can be considered for FOCUS4-C.
- If an interim CT scan is performed (recommended 8 weeks after the start of first-line treatment), it must indicate that the patient has stable or responding disease and is therefore more likely to be stable and responding by the end of first-line treatment scan.

The patient must have ample time to consider participation in FOCUS4-C and plenty of opportunities to discuss it with their clinician. If at this stage it becomes clear that the patient does not wish to, or will not be able to, travel to another site for their treatment or they are not showing enthusiasm for enrolment for FOCUS4-C, then the alternative option of FOCUS4-N should be discussed with the patient and PIS2 for FOCUS4-N (PIS2-N) provided. Where a patient is also eligible for enrolment into
another open comparison, this option should also be discussed with the patient and the relevant PIS2 provided. Please refer to the relevant molecular cohort protocol for further details.

If the site where the potential FOCUS4-C patient is registered is not treating patients in FOCUS4-C (as per their site level), the patient should be referred for consideration at a site able to deliver FOCUS4-C treatment. If the patient is willing to travel to such a site for treatment, the consent process for FOCUS4-C must continue at the randomising site and a new patient information sheet (PIS2) with the agreed site’s headed paper should be given to the patient. Following this, the patient should be ready to give or decline informed consent at the randomising site following the results of their end of first-line treatment CT scan and eligibility being checked. Once consent has been given by the patient at the randomising site, the remaining eligibility criteria assessments will be performed such as the ECG. If the patient meets all the eligibility criteria, they can be randomised as described in section 4. Obtaining final consent prior to randomisation will be the responsibility of the randomising site who will be administering treatment.

In order to investigate tumour heterogeneity and its relation to responsiveness to therapy in FOCUS4, blood samples and biopsy of accessible metastatic sites are requested prior to starting trial treatment and on progression for those patients who consent to this optional additional research. Please refer to the Master Protocol Section 4.6 and Sample Collection and Handling SOP for these optional sub-studies.

3.3 PATIENT INCLUSION AND EXCLUSION CRITERIA FOR RANDOMISATION

There will be no exceptions to eligibility requirements at the time of randomisation. Questions about eligibility criteria should be addressed prior to attempting to register or randomise the patient.

The eligibility criteria for the FOCUS4 Trial Programme have been carefully considered. The eligibility criteria are the standards used to ensure that only medically appropriate patients are considered for this trial. Patients not meeting the criteria should not join the trial. For the safety of the patients, as well as to ensure that the results of this trial can be useful for making treatment decisions regarding other patients with similar diseases, it is important that no exceptions be made to these criteria for admission to the trial.

Patients will be considered eligible for randomisation into FOCUS4-C if they fulfil all the inclusion criteria and none of the exclusion criteria as defined below.

3.3.1 INCLUSION CRITERIA

1. Registered to FOCUS4 and therefore met all the eligibility criteria for initial registration (please refer to FOCUS4 Master Protocol).
2. FFPE tumour block supplied for biomarker analysis and results reported as H3K36me3 loss on IHC, or RAS mutation and p53 mutation on gene sequencing.
3. An end of first-line treatment CT scan must have been performed no more than 28 days prior to randomisation.
4. Unidimensionally measurable disease (RECIST v1.1 classification) assessed to be stable disease, partial response or complete response after their first-line treatment.
5. Minimum 3 week gap from last chemotherapy or biological therapy administration prior to first trial drug administration.
6. WHO performance status 0 or 1 (refer to Appendix IV-C).
7. Adequate organ function within 14 days prior to randomisation:
   a) Calculated creatinine clearance >50ml/min (according to local estimation method).
   b) Serum bilirubin within normal limits or ≤1.5 x ULN in patients with liver metastases; or total bilirubin ≤3.0 x ULN with direct bilirubin within normal limits in patients with documented Gilbert’s Syndrome.
   c) ALT/AST: ≤3 x ULN (in absence of liver metastases) or ≤5 x ULN (in presence of liver metastases).
   d) Adequate haematological parameters: absolute neutrophil count (ANC) ≥1.5 x 10^9/L, haemoglobin (Hb) >9g/dL and platelets >100 x 10^9/L.
8. For women of child bearing potential, use of adequate contraceptive measures (for the definition of adequate contraceptive measures, please refer to FOCUS4 Master Protocol Appendix V for contraindications of oral contraceptives) from 2 weeks prior to starting trial treatment to one month after ending trial treatment, should not be breastfeeding and must have a negative pregnancy test within 7 days prior to randomisation. Women of non-child bearing potential should fulfil one of the following criteria at screening:
   o Post menopausal as defined by:
      − Aged ≥50 years and amenorrhoeic for at least 12 months following cessation of all exogenous hormonal treatments
      − Aged <50 years and amenorrhoeic for at least 12 months following cessation of all exogenous hormonal treatments and with luteinising hormone and follicular stimulating hormone levels in the post-menopausal range
   o Documentation of irreversible surgical sterilisation by hysterectomy, and/or bilateral oophorectomy and/or bilateral salpingectomy but excludes bilateral tubal ligation
9. Male patients should be willing to abstain or use barrier contraception (i.e., condoms) for the duration of the trial treatment and for 3 months after stopping AZD1775. Male patients wishing to father children whilst taking AZD1775 and 3 months after stopping trial treatment should be counselled on freezing sperm samples before randomisation.
10. Patients should have sufficient capacity for informed consent.
11. Written informed consent for randomisation to FOCUS4-C.

3.3.2 EXCLUSION CRITERIA
1. Grade >1 toxicity from prior therapy (except alopecia or anorexia).
2. Inability to swallow oral medications. Note: Patient may not have a percutaneous endoscopic gastrostomy (PEG) tube or be receiving total parenteral nutrition (TPN).
3. Serious active infection at the time of trial entry, or another serious underlying medical condition that would impair the ability of the patient to receive trial treatment.
4. Mean resting corrected QTc interval using the Fridericia formula (QTcF) >450 msec/male and >470 msec/female (as calculated per institutional standards) obtained from 3
electrocardiograms (ECGs) 2-5 minutes apart at randomisation, or congenital long QT syndrome.

5. Known uncontrolled or symptomatic angina, arrhythmias or congestive cardiac failure, evidence of transmural infarction on ECG, poorly controlled hypertension (systolic >180mmHg or diastolic >100mmHg), significant valvular disease or history of high risk dysrhythmia (such as ventricular tachycardia or ventricular fibrillation [includes ventricular triplets]).

6. AZD1775 should not be given to patients who have a history of Torsades de pointes unless all risk factors that contributed to Torsades have been corrected. AZD1775 has not been studied in patients with ventricular arrhythmias or recent myocardial infarction.

7. Active or previous peptic ulceration unless well-controlled on PPIs for at least 6 months or resolved with HP eradication.

8. Previous gastrointestinal bleeding except where the cause of the bleeding has been surgically removed.

9. Major surgical procedures ≤28 days of beginning trial treatment, or minor surgical procedures ≤7 days. No waiting period required following port-a-cath or other central venous access placement.

10. Concomitant use of products known to be sensitive to CYP3A4 substrates or CYP3A4 substrates with a narrow therapeutic index, or to be moderate to strong inhibitors/inducers of CYP3A4 which cannot be discontinued 2 weeks prior to Day 1 of AZD1775 and withheld throughout the trial until 2 weeks after the last dose of trial drug (these drugs are contraindicated – please refer to Appendix IX-C for full list).

11. Unable to stop or substitute other prohibited drugs, most notably aprepitant, fosaprepitant, atorvastatin, rosuvastatin, simvastatin, lovastatin (starting 2 weeks prior to trial treatment) and all herbal remedies (1 week prior to starting trial treatment other than St. John’s wort which must be stopped 3 weeks prior to starting trial treatment). For further advice on common prescribed prohibited medications please refer to section 5.9, and for a list of prohibited medication please refer to Appendix IX-C.

12. Unwillingness or inability to follow the procedures outlined in the protocol.

13. Known HIV, hepatitis B, or hepatitis C infection. Patients who have evidence of clearance of hepatitis B infection can only be randomised with approval of the FOCUS4-C Co-Chief Investigators or clinical member of the TMG.

### 3.4 ACTIONS PRIOR TO RANDOMISATION

Once the biomarker panel results have been received at the MRC CTU at UCL, the trial team will notify the registering site if the patient should be considered for FOCUS4-C. If the interim CT scan has shown evidence of stable or responding disease, and the patient does not meet any of the exclusion criteria as documented in Section 3.3, the site can then give the patient the FOCUS4-C PIS2 and discuss the details of the relevant randomisations with the patient including, if required, whether they need to be referred to a randomising site. In particular, consideration must be given in advance as to whether the patient currently takes any of the listed prohibited medications (Appendix IX-C): patients will only be eligible for FOCUS4-C if they can stop the medication,
substituting for an alternative agent if necessary. If the patient is found not to be eligible for FOCUS4-C or preliminary discussions with the patient have suggested that the patient may not wish to enter FOCUS4-C, the patient must be offered entry into FOCUS4-N instead. Please refer to the FOCUS4-N Protocol for further information on this comparison and its processes. Where a patient is also eligible for enrolment into another open comparison, this option should also be discussed with the patient and the relevant PIS2 provided.

If the patient does express an interest in participating in FOCUS4-C, any sites ineligible for randomisation must refer the patient to a randomising site for further eligibility and consenting processes. Once the patient is in the care of the randomising site, the site must consider fully the patient’s eligibility according to the list provided under Section 3.3. If they meet all of the eligibility criteria determined by routine hospital tests, then they should be consented to join FOCUS4-C and sign the consent form for FOCUS4-C (CF2-C). For non-routine tests such as an ECG, these should only be performed once the patient has consented to join FOCUS4-C. If they are subsequently found to be ineligible from those test findings, they will not be able to enter FOCUS4-C and the local clinician must explain to the patient why the latest tests have made them ineligible.

**Action steps for randomising sites before the patient is randomised:**
- Ensure the End of Registration eCRF has been completed.
- Confirm the patient’s general fitness for treatment.
- Check all inclusion and exclusion criteria in Section 3.3 and confirm the patient is fully eligible in light of all baseline investigations, including WHO status (see Appendix IV-C).
- Ensure patient is not currently taking any prohibited medications.
- Check full blood count and biochemistry. Calculated creatinine clearance using local methods. If the creatinine clearance is $<50$ ml/min, a formal method of determining GFR is required using local methods.
- Ensure an ECG is performed within 14 days prior to randomisation.
- Check the patient has documented measurable disease (RECIST v1.1 criteria, Appendix V-C) stating SD, PR or CR after end of first-line treatment CT scan. This scan must be **within 28 days prior to randomisation**.
- Ensure that the patient understands that they are free to give or withhold permission for the use of samples for the additional bowel cancer research and other sub-studies without affecting their participation in the trial.
4 SELECTION OF PATIENTS FOR FOCUS4-C RANDOMISATION

4.1 RANDOMISATION

Prior to randomisation, the FOCUS4-C eligibility checklist and section A of the Randomisation Form must be completed documenting the clinical findings as well as the results of the end of registration CT scan. This scan will become the baseline scan upon which all subsequent CT scans are compared in the trial. If the patient has given consent for the optional additional blood samples (question 6 on the consent form), these samples must be taken. If the patient has given consent to have the optional additional biopsy, this procedure should be scheduled prior to the start of the molecular comparison therapy (please refer to Section 4.6 of the Master Protocol and Sample Collection and Handling SOP).

It is requested that the Randomisation Form is completed before calling the randomisation line:

**RANDOMISATIONS**
To randomise, call MRC CTU at UCL, Monday to Friday 9 am to 5 pm (except on Bank holidays)
Tel: 020 7670 4777

At randomisation the original trial number will be appended with the letter of the comparison the patient has entered (C for FOCUS4-C). This full trial number must be used for all subsequent documentation throughout the trial.

Further details on the process of randomisation can be found in Section 9.1 - Method of Randomisation.

4.2 RANDOMISATION CODES

FOCUS4-C is not blinded. Patient and clinicians will be aware of the allocation to Active Monitoring or AZD1775.

4.3 TRANSLATIONAL RESEARCH SAMPLE COLLECTION

In order to investigate tumour heterogeneity and its relation to responsiveness to therapy in the FOCUS4 Trial Programme, blood samples and biopsy of accessible metastatic sites are requested prior to starting trial treatment and on progression for those patients who consent to this additional research. This is very important work required to understand and optimise patient selection and to help understand mechanisms of resistance. We expect most patients to be willing, if asked, to provide extra samples for this work and investigators are strongly encouraged to approach patients for their consent to these samples. These samples are optional so the patient has freedom to refuse
the collection of the additional samples and can still enter the trial. Please see Section 4.6 of the Master Protocol and Sample collection and Handling SOP for these optional sub-studies.

4.4 CO-ENROLMENT GUIDELINES

Patients involved in other clinical trials that relate to their colorectal cancer are unlikely to be eligible for entry into FOCUS4-C (see section 4.7 of the Master Protocol - Co-enrolment guidelines). At the time of writing this protocol, the investigators are not aware of any other open trials that will conflict with FOCUS4-C.

4.5 REGISTERED PATIENTS WHO DO NOT CONSENT TO RANDOMISATION

Patients may register in FOCUS4 but then decide not to be randomised in any subsequent comparisons that they are offered. An End of Registration eCRF should be completed for all such patients, with reasons for their refusal documented wherever possible.
5 TREATMENT OF PATIENTS IN FOCUS4-C

5.1 INTRODUCTION AND TREATMENT ALLOCATION

FOCUS4-C is an open-label, randomised controlled comparison. Once patients have consented to FOCUS4-C, they will be randomly assigned to one of the following treatment arms in a 2:1 ratio in favour of the AZD1775:

- **Arm C1**: Active Monitoring (one third allocation)
- **Arm C2**: AZD1775 (two thirds allocation)

PIS2-C, FOCUS4-C diary card and the consent process will give further information relating to the administration and side effects of AZD1775. Patients may also be offered the opportunity to take part in additional studies as part of this trial including biopsy of metastatic lesions and blood samples (please refer to Section 4.6 of the Master Protocol and Sample collection and Handling SOP).

Trial treatment should start as soon as possible after randomisation subject to a minimum wash out period of 3 weeks from end of first-line treatment (any chemotherapy or biological therapy administration). Please note that patients will be regarded as off treatment if they have not started their trial treatment more than 28 days after the end of their 3 week wash out period (Therefore the maximum time allowed from finishing first-line treatment to starting trial treatment is 7 weeks).

Please refer to Appendix VI-C for toxicity and dose modifications. It is the responsibility of the treating consultant to ensure the treatment regimens are followed, in particular dose modifications should only be made after consulting this written protocol and Appendix VI-C (if in doubt, please discuss with the MRC CTU at UCL).

5.2 TREATMENT AND CT SCAN SCHEDULE

5.2.1 ACTIVE MONITORING ARM

No chemotherapy or biological treatment should be given to patients in the active monitoring arm. Patients require CT scans every 8 weeks from randomisation and full reporting of safety outcomes. Clinical evaluation and assessment of toxicities will need to be performed every 3-4 weeks by a research nurse or doctor as appropriately delegated and a Toxicity and Symptom eCRF completed. All patients will be evaluated with a CT scan every 8 weeks when a Progress eCRF is also required (see Trial Assessment schedule in Section 6).

On disease progression, patients should restart their first-line treatment or move onto a standard second-line therapy at the oncologist’s discretion. After progression, a Progress eCRF must be completed at 3 months and then every 6 months until patient death or the end of the trial.

5.2.2 AZD1775 ARM

The starting dose for AZD1775 is 250mg once daily for 5 consecutive days during weeks 1 and 2 of a 3-week cycle (250mg O.D., d1-5 & 8-12, q21d). Thus a total of 10 doses of AZD1775 will be taken in each 21-day cycle. The AZD1775 dose is not adjusted for patient size. A formal tolerability review
will be conducted 42 days after the 12th patient has initiated AZD1775 treatment, and a higher starting dose of 300mg O.D., d1-5 & 8-12, q21d will be considered. In this event all participating centres will be notified of the dose change, and all new patients will be started at the 300mg dose.

Regardless of the starting dose, patients who require a dose reduction for toxicity will be prescribed AZD1775 200mg O.D., d1-5 & 8-12, q21d. Please refer to Appendix VI-C for information about Toxicity and Dose Modifications.

AZD1775 is supplied in formulations of 100mg and 25mg capsules. The 250mg dose is therefore made up of 2 x 100mg capsules and 2 x 25mg capsules; the 300mg dose is made up of 3 x 100mg capsules, and the 200mg dose is made up of 2 x 100mg capsules. Separate bottles will be supplied for the 100mg capsules and the 25mg capsules. Sites will be provided with some surplus bottles of each formulation. The 100mg bottle will have a blue label and the 25mg a white label to aid differentiation for the patient.

<table>
<thead>
<tr>
<th>Cycle day</th>
<th>1</th>
<th>2</th>
<th>3</th>
<th>4</th>
<th>5</th>
<th>6</th>
<th>7</th>
<th>8</th>
<th>9</th>
<th>10</th>
<th>11</th>
<th>12</th>
<th>13</th>
<th>14</th>
<th>15</th>
<th>16</th>
<th>17</th>
<th>18</th>
<th>19</th>
<th>20</th>
<th>21</th>
</tr>
</thead>
<tbody>
<tr>
<td>AM</td>
<td>✓</td>
<td>✓</td>
<td>✓</td>
<td>✓</td>
<td>✓</td>
<td>-</td>
<td>-</td>
<td>✓</td>
<td>✓</td>
<td>✓</td>
<td>✓</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>-</td>
</tr>
</tbody>
</table>

✓ = Dose to be taken  
- = No dose required

AZD1775 will be self-administered and should be taken at the same time each day it is required, either two hours before or two hours after food. If a patient misses a dose this should be taken as soon as possible, but not more than eight hours after the missed scheduled dose. If vomiting occurs after taking AZD1775, the patient should be instructed not to retake the capsules but to wait and take the next scheduled dose of AZD1775. Patient reported adherence to trial treatment will be collected using diary cards. A description of how to take AZD1775, including what to do if doses are missed is provided in PIS2-C and the FOCUS4-C diary card.

Due to high incidence of nausea and vomiting observed in previous studies, patients randomised to receive AZD1775 must receive the following:

**5HT3 Antagonist:** Patients should routinely receive an oral 5HT3 antagonist (e.g. ondansetron 8 mg or granisetron 1 mg) with each dose of the study drug. A supply of 5HT3 antagonist should also be provided for PRN use after the AZD1775 dosing period.

**Corticosteroid:** Patients should routinely receive oral dexamethasone 4 mg O.D. on the first day of each AZD1775 dosing period (d1 and d8 of each cycle), unless steroid use is contraindicated or poorly tolerated. At clinical discretion, dexamethasone may be continued on the second to fifth days (d2-5 and d9-12) of AZD1775 dosing, potentially at a lower dose to limit steroid side-effects.

Additional anti-emetics may be added to this basic regimen, and doses adjusted, at any point as clinically indicated. Please note, however, that aprepitant/fosaprepitant use is not permitted because of potential interaction with AZD1775.
No additional anticancer chemotherapy or biological treatment may be given to patients in either trial arm.

If a patient is unable to tolerate treatment at Day 8 (due to ongoing AZD1775 toxicity or any other reason) then the day 8-12 doses should be omitted: these doses should only be given as per the cycle schedule and therefore should not be delayed. Assessment for treatment for the next cycle should occur as scheduled and AZD1775 dose reduction may be considered (see Appendix VI-C). When the next cycle is due, if the patient is unable to restart treatment due to ongoing AZD1775 toxicity or for any other reason, a maximum of 21 days delay is allowed for resolution. If a cycle is delayed for any reason, for longer than 21 days from scheduled Day 1, the patient is considered off-treatment and this must be documented on the Progress eCRF. Please refer to Appendix VI-C for toxicity and dose modifications.

All patients in the AZD1775 arm will be evaluated with a CT scan every 7-9 weeks from randomisation. Toxicities and symptoms will be assessed every 3 weeks from start of trial treatment.

Progression-free survival (PFS) is the primary outcome measure for FOCUS4-C and therefore clinicians are required to use a consistent approach to treatment duration. Treatment should be continued until progressive disease is identified on radiological grounds (RECIST v1.1) (Appendix V-C) or the development of cumulative toxicity or because of patient choice to stop treatment. When this occurs, patients will be considered off-treatment (but still in the trial) and may restart their first-line treatment at their clinician’s discretion. For patients discontinuing treatment with AZD1775 there should be at least a 2 week washout period between the last dose before restarting first-line chemotherapy and that there is resolution of toxicity to Grade ≤1.

However, please note that patients who discontinue trial drug for reasons other than objective disease progression should be followed with tumour assessments every 7-9 weeks until objective disease progression by RECIST v1.1 even if they have started subsequent anti-cancer therapies. Toxicity assessment should also continue until disease progression is reported.

5.2.3 TOLERABILITY PHASE AND PLANNED REVIEW OF AZD1775 STARTING DOSE

At the time of initiating FOCUS4-C, ongoing studies have been reviewed to select the optimum dose of AZD1775 to ensure good tolerability whilst achieving biologically active drug levels. Based on current safety and pharmacokinetic data, the starting dose of 250mg O.D. d1-5 & 8-12, q21d has been selected. Ongoing studies elsewhere are exploring a higher dose of 300mg O.D.

Therefore, FOCUS4-C will include a planned independent tolerability review and an opportunity to increase the starting dose to 300mg O.D. d1-5 & 8-12, q21d. The review will be performed by the IDMC, and will be scheduled to take place at least 42 days after the 12th patient has started AZD1775 treatment. The IDMC will be asked to consider all data from the FOCUS4-C trial, alongside up-to-date data from all relevant trials elsewhere (to be provided in confidence by AstraZeneca).

The IDMC will recommend, on the basis of their review:
(a) increase to 300mg O.D. d1-5 & 8-12, q21d
or  (b) continue the study at 250mg O.D. d1-5 & 8-12, q21d
or  (c) a reduction in starting dose.

The IDMC decision will not be constrained; however, as guidance, escalation to 300mg will be considered if \( \geq 9 \) of 12 patients have tolerated two full cycles without requiring dose reduction, and provided any external data provided by AstraZeneca supports this. Conversely, a lower starting dose (e.g. 200 mg O.D.) may be recommended if fewer than 6 of 12 patients tolerated 250mg for two cycles.

The IDMC will also be invited to comment on the suitability of extending FOCUS4-C treatment to a wider range of FOCUS4 sites, and on the need and timing for further tolerability reviews.

5.2.4 DOSE MODIFICATIONS, INTERRUPTIONS & DISCONTINUATIONS FOR THE AZD1775 ARM

- Based on the safety data from the completed AZD1775 clinical studies and preliminary data from ongoing studies-adverse drug reactions to AZD1775 monotherapy include: anaemia, neutropenia, thrombocytopenia, QTc prolongation, gastrointestinal events such as dyspepsia, diarrhoea, nausea and vomiting (with or without dehydration or serum electrolyte decreases), as well as decreased appetite.
- Based on information emerging during the clinical development programme of AZD1775, potential risks with AZD1775 monotherapy include asthenia/fatigue, febrile neutropenia, gastrointestinal haemorrhage, lymphopenia/lymphocyte count decreased, leukopenia/WBC count decreased, myalgia, stomatitis, sepsis and transaminases elevation.

Grade \( \geq 3 \) toxicities (assessed utilising the CTCAE v4) or persistent toxicities which are considered medically significant or not tolerated by the patient necessitate stopping treatment with AZD1775 until resolution to Grade \( \leq 1 \). Dose reductions or holds and initiation of supportive care are allowed as clinically indicated by the treating physician. One dose reduction of AZD1775 will be allowed: regardless of whether the starting dose was 250 mg or 300mg, patients requiring a dose reduction should receive AZD1775 200mg O.D., d1-5 & 8-12, q21d. Dose re-escalation is not permitted.

Specific advice and dose modifications for these can be found in Appendix VI-C.

5.3 PROTOCOL TREATMENT DISCONTINUATION

In consenting to FOCUS4-C, patients are consenting to trial treatment, follow-up and data collection. However, an individual patient may stop treatment early for any of the following reasons:

- Disease progression
- Unacceptable cumulative toxicity or adverse event
- Intercurrent illness that prevents further treatment
- Pregnancy
- Any change in the patient’s condition that justifies the discontinuation of treatment in the clinician’s opinion
• Inadequate compliance with the protocol treatment in the judgement of the treating physician
• Withdrawal of consent for treatment by the patient

Patients who discontinue trial drug for reasons other than objective disease progression should be followed with regular tumour assessments (7-9 weekly) until objective disease progression by RECIST 1.1 even if they started subsequent anti-cancer therapies. Toxicity and symptom assessment should occur within 4 weeks of end of treatment and then continue 7-9 weekly alongside tumour assessments until disease progression is reported. For patients discontinuing treatment with AZD1775 there should be at least a 2 week washout period between the last dose before restarting first line chemotherapy and that there is resolution of toxicity to Grade ≤1.

As the patient’s participation in the trial is entirely voluntary, they may choose to discontinue the trial treatment at any time without penalty or loss of benefits to which they are otherwise entitled. Although the patient is not required to give a reason for discontinuing their trial treatment, a reasonable effort should be made to establish this reason while fully respecting the patient’s rights.

Patients should remain in the trial for the purpose of follow-up and data analysis (unless the patient withdraws their consent from all parts of the trial). Data will be kept and included for patients who stop follow-up early, unless a patient is withdrawn from follow-up. For information on withdrawals please refer to Section 6.6 - Early Stopping of Follow-up.

5.4 COMPLIANCE & ADHERENCE

Diary cards will be supplied for patients to complete for each cycle. Patients will document when each dose is taken and any missed doses (with reasons). This information will be checked by the research nurse at each 3 weekly visit and summarised on the Treatment eCRF.

5.5 TREATMENT DATA COLLECTION

Trial treatment must be recorded on the Treatment eCRF. In addition, reasons for any dose delays, reductions or omissions or for permanent discontinuation of trial treatment must be documented in the appropriate part of the CRF.

The number of doses taken will be recorded by the patient on the diary card that they complete on a daily basis, documenting any missed doses (with reasons) as per in section 5.4. All the packaging and unused capsules should be returned by patients and returned and stored in pharmacy. At each 3 weekly clinical appointment, the research nurse will review and summarise the patient’s drug compliance. This summary will be documented on the Treatment eCRF.

5.6 TREATMENT ON PROGRESSION

The principle of intermittent therapy, as used in OPTIMOX2[36] and COIN[37], is that patients are expected to restart their first-line therapy on evidence of progression of their metastatic or locally advanced disease. The higher the level of reintroduction of first-line therapy the better the outcome
appeared to be in the OPTIMOX1[38] and 2 analyses, for oxaliplatin based first-line therapy. Therefore it is expected that all patients in FOCUS4-C will restart on their first-line therapy on progression, if a reasonable time has passed since last administration. This reintroduction of chemotherapy is at the clinician’s discretion as they assess what is to be in the patient’s best interests. For patients discontinuing treatment with AZD1775 there should be at least a 2 week washout period between the last dose before restarting first line chemotherapy and that there is resolution of toxicity to Grade ≤1. Reasons for not restarting on the same first-line therapy will be requested on the Progress eCRF and may include:

- Clinical deterioration or an intercurrent illness preventing further treatment
- Residual cumulative toxicity (notably oxaliplatin induced neuropathy) which prevents restarting the same schedule
- Very early disease progression after stable disease on first-line therapy: this is an issue of clinical judgement.

Data will be collected about subsequent chemotherapy utilisation. Follow-up after progression will be collected at 3 months and then every 6 months until patient death or the end of the trial.

5.7 AZD1775

5.7.1 PRODUCTS

AZD1775 will be supplied by AstraZeneca and labelled and distributed by Fisher Clinical Services. Trial drug will be packaged and labelled in accordance with local regulations and Good Manufacturing Practice, stating that the drug is for clinical trial use only and should be kept out of the reach and sight of children. AZD1775 should be kept in a secure place under appropriate storage conditions (store below 30°C). AZD1775 will be supplied as 100mg and 25mg capsules, in 75cc HDPE bottles with 20 capsules per bottle. For further information on AZD1775, please refer to the AZD1775 Investigator Brochure.

5.7.2 DISPENSING AND DRUG SUPPLIES

- An initial supply of AZD1775 will be sent to the randomising site when a potential FOCUS4-C patient is identified. This will be ordered by MRC CTU at UCL.
- Further resupply of AZD1775 should be ordered from Fisher Clinical Services using the Drug Order Form. Please refer to the Pharmacy SOP for further information.
- Trial medication will be dispensed on a 3 week cycle.
- For each cycle, patients will be supplied with sufficient medication to last them until their next visit.
- All patients randomised to receive AZD1775 should routinely receive an oral 5HT3 antagonist (e.g. ondansetron 8mg or granisetron 1mg) with each dose of the study drug. A supply of 5HT3 antagonist should also be provided for PRN use after the AZD1775 dosing period. In addition, patients should routinely receive oral dexamethasone 4mg O.D. on the first day of each AZD1775 dosing period (i.e. d1 and d8 of each cycle), unless steroid use is contraindicated or poorly tolerated. At clinical discretion, dexamethasone may be continued on the remaining days of
AZD1775 dosing (d2-5 and d9-12 of each cycle), potentially at a lower dose to limit steroid side-effects. Additional anti-emetics may be added to this basic regimen, and doses adjusted, at any point as clinically indicated. Please note, however, that aprepitant/fosaprepitant use is not permitted because of potential interaction with AZD1775.

- Supportive medications for the toxicities as listed in 5.2.3 are specified in Appendix VI-C.
- Accountability logs for AZD1775 will be provided within the Pharmacy SOP. Please refer to section 5.7.3 below for further details.
- The guidelines in this protocol are in line with the manufacturer’s recommendations at the time of writing.

5.7.3 ACCOUNTABILITY & UNUSED DRUGS

- The trial pharmacist will sign a document to confirm that local hospital systems are in place to cover drug ordering, drug receipt, drug storage and dispensing, and their systems will enable accurate traceability of AZD1775.
- Full drug accountability records must be maintained for AZD1775 which will be provided in the Pharmacy SOP at site activation. All AZD1775 receipts, dispensing, patient returns and destructions must be recorded on the logs provided.
- Each patient’s accountability records should be submitted to the MRC CTU at UCL when each patient discontinues trial treatment. These may be used to measure overall patient drug compliance.
- Details of how to dispose of expired or unused AZD1775 will be given in the Pharmacy SOP. Any unused capsules are to be returned to pharmacy for disposal. Used or partially used packets of drug should be disposed of at site according to local policy and recorded on the FOCUS4-C destruction log and accountability logs.

5.8 OVERDOSE OF TRIAL MEDICATION

A dose of AZD1775 in excess of that specified according to the protocol will constitute an overdose. There is currently no antidote to AZD1775 and the treatment of overdose should be supportive for the underlying symptoms. To date, there has been one patient who has experienced an overdose with AZD1775 which was associated with adverse events.

For overdoses associated with SAE, standard reporting timelines apply. For other overdoses not associated with an SAE, data should be recorded on the Treatment and Toxicity & Symptoms eCRFs.

5.9 MEDICATIONS OUTSIDE OF THE TRIAL

Concomitant use of products known to be sensitive to CYP3A4 substrates or CYP3A4 substrates with a narrow therapeutic index, or to be moderate to strong inhibitors/inducers of CYP3A4 which cannot be discontinued 2 weeks prior to Day 1 of AZD1775 and withheld throughout the trial until 2 weeks after the last dose of trial drug are not permitted.
All prohibited medication must be discontinued two weeks prior to starting trial treatment (all herbal remedies must be discontinued one week prior to starting trial treatment other than St. John’s wort which must be stopped 3 weeks prior to starting trial treatment.

5.9.1 PERMITTED AND PROHIBITED CONCOMITANT MEDICATIONS

Patients requiring therapeutic warfarin or coumarin-derivative anticoagulants will be monitored with international normalized ratio (INR) and prothrombin time (PT) as clinically indicated. Low molecular weight heparin (LMWH), rivaroxaban, or equivalent anticoagulant therapy is permitted where clinically indicated.

Transporter studies have demonstrated that AZD1775 is an inhibitor of BCRP, so may potentially interact with other drugs affected by BCRP-mediated efflux, notably some statins including rosuvastatin. Patients on statins who are randomised to AZD1775 should normally be asked to stop taking the statin while on trial treatment. In the unlikely event that continuation of a statin is considered clinically essential, please use the minimum effective dose of atorvastatin/simvastatin/lovastatin and notify this decision to the MRC CTU at UCL. Patients may restart their statins at their pre-study dose after completing trial treatment subject to a 1 month wash out.

Please refer to Appendix IX-C for a list of medication that must be avoided due to potential drug interactions with AZD1775. Please note that this includes (but is not limited to) the following commonly used drugs:

- Aprepitant/ fosaprepitant
- Atorvastatin
- Simvastatin
- Lovastatin
- Rosuvastatin
- Clarithromycin (but topical use for acne is acceptable)
- Erythromycin (but topical use for acne is acceptable)
- Fluconazole
- Ketoconazole
- Diltiazem
- Verapamil
- Amiodarone
- Carbamazepine
- Phenobarbitones
- Phenytoin
- St John’s Wort and many other herbal medications

Please note that grapefruit and Seville oranges (and other products containing Seville oranges, including marmalade) should not be consumed during AZD1775 treatment.

5.9.2 MEDICATIONS AND FOOD TO BE USED WITH CAUTION

- Digoxin (levels should be monitored according to local practice).
- Metformin should be used with caution in this study as recent in vitro transporter data have shown AZD1775 is an inhibitor of Multidrug and Toxin Extruder 1 (MATE1) and MATE2K.
- Non-steroidal anti-inflammatory drugs should be used with caution in this study due to the small increased risk of GI haemorrhage and co-prescription of dexamethasone with AZD1775 treatment. When necessary we would recommend that a proton pump inhibitor should be co-prescribed.

For a full list of medications to be avoided or used with caution in combination with AZD1775 see Appendix IX-C.
## 6 TRIAL ASSESSMENTS & FOLLOW-UP FOR FOCUS4-C

### Table 6.1: FOCUS4-C Assessment Schedule
(The table is also provided in the Trial Summary on page viii)

<table>
<thead>
<tr>
<th></th>
<th>Pre-randomisation</th>
<th>Every 3-4 weeks until end of trial treatment *</th>
<th>Every 7-9 weeks until progression</th>
<th>If any SAE occurs</th>
<th>Within 4 weeks of end of trial treatment (progression, toxicity or patient choice)</th>
<th>Follow-up after progression at 3 months and then 6 monthly</th>
</tr>
</thead>
<tbody>
<tr>
<td>Clinical evaluation</td>
<td>X^</td>
<td>X</td>
<td>X</td>
<td>X</td>
<td>X</td>
<td>X</td>
</tr>
<tr>
<td>(including Blood pressure, Pulse rate, weight measurement)</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>WHO PS</td>
<td>X^</td>
<td>X</td>
<td>X</td>
<td>X</td>
<td>X</td>
<td>X</td>
</tr>
<tr>
<td>Pregnancy Test (for women of child bearing potential)^~</td>
<td>X^</td>
<td>X</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Toxicity (NCI CTCAE)</td>
<td>X^</td>
<td>X</td>
<td>X</td>
<td>X</td>
<td>X</td>
<td></td>
</tr>
<tr>
<td>Review use of concomitant medications</td>
<td>X^</td>
<td>X</td>
<td>X</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>FBC, U&amp;Es, LFTs, ALT/AST, ALKP</td>
<td>X^</td>
<td>X</td>
<td>X</td>
<td>X</td>
<td>X</td>
<td></td>
</tr>
<tr>
<td>CEA, LDH</td>
<td>X^</td>
<td>X</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>GFR</td>
<td>X^†</td>
<td>X</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>CT Scan</td>
<td>X¶</td>
<td></td>
<td>X</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>RECIST v1.1 response from CT scan</td>
<td>X¶</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>ECG</td>
<td>X^</td>
<td>X</td>
<td>X</td>
<td></td>
<td>X</td>
<td>X α</td>
</tr>
<tr>
<td>Dispensing AZD1775, Diary Card assessment</td>
<td>X∞#</td>
<td>X</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>CRFs/eCRFs required</td>
<td></td>
<td>Randomisation CRF</td>
<td>Treatment eCRF &amp; Toxicity &amp; Symptom eCRF</td>
<td>Progress eCRF &amp; Toxicity &amp; Symptom eCRF</td>
<td>SAE Report CRF</td>
<td>Toxicity &amp; Symptom eCRF</td>
</tr>
<tr>
<td>Optional assessments (If specific consent given)</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Circulating free DNA</td>
<td>X‡</td>
<td>X</td>
<td></td>
<td>X‡ for progression only</td>
<td>X$</td>
<td></td>
</tr>
<tr>
<td>Optional Tumour biopsy</td>
<td>X$</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

**Key for symbols**
- ^: Within 14 days prior to randomisation
- †: Not required if calculated creatinine clearance >50ml/min (using local protocols)
- ¶: Pre-randomisation CT (baseline comparator scan) within 28 days prior to randomisation
- #: For patients on Arm C2 (AZD1775) only. ECG to be performed within 1 week prior to starting each cycle while on AZD1775 but not required prior to cycle 1 as pre-randomisation ECG is available.
· Women of child bearing potential on Arm C2 (AZD1775) must have a negative pregnancy test 7 days prior to randomisation and every 3 weeks while on trial treatment. They must also be using adequate contraception for two weeks prior to starting trial treatment, while receiving AZD1775 and for one month following end of trial treatment. Male patients should be willing to abstain or use barrier contraception (i.e., condoms) for the duration of the trial treatment and for 3 months after stopping AZD1775.

* Patients in the active monitoring arm still require appointments with the Research Nurse every 4 weeks to check they are well.

α If clinically indicated by the event

∞ Until patient stops trial treatment due to progression, cumulative toxicity or patient choice

‡ Optional blood samples not required if patient withholds consent for molecular research

§ Optional tumour biopsies not required if patient withholds consent to additional biopsies. If patient consents to this sub-study, biopsy should be taken after randomisation (prior to starting trial treatment) and on progression.

6.1 CASE REPORT FORM (CRF) RETURN GUIDELINES

Please also refer to Table 6.1.

· End of Registration eCRF should be completed prior to randomisation by the registering site on the registration MACRO database.

· Randomisation Eligibility Checklist - the randomisation eligibility checklist must be signed by the randomising clinician prior to randomisation and submitted to the MRC CTU at UCL. Please note the patient will not be created in the FOCUS4 Main Database until this form is received at the MRC CTU at UCL. The FOCUS4 team will inform the site when this has occurred.

· Section A of the Randomisation CRF should be completed before the patient is randomised as it contains the questions required to randomise the patient. Once the patient is created by FOCUS4 Team in the FOCUS4 Main Database, this must be inputted as soon as possible.

· Section B of Randomisation CRF should be entered into the FOCUS4 Main Database with section A as soon as possible when the patient has been created.

· Treatment eCRFs for all cycles for patients in Arm C2

· Toxicity and Symptom eCRF is required every 3-4 weeks

· Progress eCRF are due every 8 weeks until progression and must be submitted within one month of being due. Following progression it is due at 3 months and then 6 monthly thereafter progression until death.

· SAE Report CRF must be completed for each SAE and sent to the MRC CTU at UCL according to the timelines described in Figure 7.1. These will be paper CRFs.

Please refer to the FOCUS4 Data Provision Guidelines for further details.

6.2 PROCEDURES FOR ASSESSING EFFICACY

· During the registration period, CT scans are required at the start and finish of standard first-line treatment with an interim scan (recommended at 8 weeks). Please refer to the Master
Protocol. The end of treatment CT scan prior to randomisation will be the baseline comparator scan for FOCUS4-C.

- Once patients enter FOCUS4-C, they will have CT scans every 7-9 weeks from randomisation until progression.
- **IMPORTANT NOTE:** The FOCUS4 trial is investigating efficacy in terms of repeated measurements from CT scans. Therefore the local investigators must consider the CT scanning facilities in their referral area and ensure that the scanners used throughout the patient’s follow-up period, provide consistent and comparable measurements. Ideally all CT scans should be performed using the same scanner but some flexibility can exist if different scanners are known to have very similar specifications and scanning protocols and radiologists in all sites can confidently report response according to RECIST v1.1 criteria.
- RECIST v1.1 will be used for assessment, all measurements should be captured on the Progress Form.

### 6.3 PROCEDURES FOR ASSESSING SAFETY

- All toxicities should be assessed, graded, recorded on clinical records and eCRFs and appropriately managed at each consultation using CTCAE version 4 until disease progression (refer to FOCUS4-C Assessment schedule).
- Specific information for safety reporting is documented in Section 7 of this protocol.

### 6.4 PROCEDURES FOR ASSESSING QUALITY OF LIFE

Currently there is no assessment of QoL in FOCUS4-C. If FOCUS4-C does proceed to a phase 3 trial (stage III), QoL may be formally assessed utilising EQSD. Sites will be notified and assessments schedules updated accordingly as a protocol amendment.

### 6.5 OTHER ASSESSMENTS

For patients who have consented to the optional blood sample collection for circulating DNA, these must be collected every 8 weeks and within 4 weeks of disease progression. Please refer to Section 4.6 of the Master Protocol for further details.

Optional biopsies at randomisation and on progression will be performed for patients in both arms of FOCUS4-C. Treating sites with appropriate facilities will be eligible to take part in these sub-studies, please refer to Section 4.6 in the Master Protocol and the Sample Collection and Handling SOP for further details. PIS3 and CF3 are to be utilised to consent patients to this sub-study.

### 6.6 EARLY STOPPING OF FOLLOW-UP

If a patient chooses to discontinue their trial treatment, they should always be followed up providing they are willing, that is, they should be encouraged to not leave the whole trial. However, their decision must be respected and the patient may stop follow up early from FOCUS4 as requested. The MRC CTU at UCL should be informed of this in writing.
Patients will be followed up in the long-term through usual mechanisms, which may include flagging with the NHS Digital, or similar approaches unless consent is withdrawn to this.

### 6.7 PATIENT REFERRALS AND TRANSFERS

It is planned to allow Level 2 and Level 3 sites to randomise and treat in FOCUS4-C; however, during the initial tolerability phase (18 patients to be randomised) only Level 3 sites will be permitted to do so.

At FOCUS4 sites which are not randomising patients into FOCUS4-C, potentially eligible patients may be approached and the informed consent process initiated, so that they can decide whether they wish to be referred to a participating site to allow them to be consented, randomised and treated; however, final consent and randomisation must be performed at the randomising site. Once referral has occurred, the randomising site will take over responsibility for the patient in FOCUS4; until this has been done, responsibility for the patient in FOCUS4 lies with the original registering site.

After randomisation, following completion of treatment, the patient may be transferred back to the original registering site. Permission must be obtained from both sites and all outstanding data and queries resolved. The patient will need to sign a new consent form with original hospital headed paper. Only once all this has been done, the MRC CTU at UCL will transfer access to the patient’s eCRFs to the new site. Then the original site will take over responsibility for the patient’s participation in FOCUS4. Until this has been done, responsibility for the patient’s participation in FOCUS4 lies with the treatment site.

If a patient moves residence from the area, every effort should be made for the patient to be seen at another participating FOCUS4 trial site of the required level. The patient will need to sign a new consent form with the new hospital headed paper and all outstanding data and queries must be resolved. Once this has been done, the new site will take over responsibility in FOCUS4 for the patient; until this has been done, responsibility for the patient in FOCUS4 lies with the original site. The MRC CTU at UCL will then transfer over access to the patient’s eCRFs to the new site. A Patient Transfer confirmation form must be completed by both sites and sent to the MRC CTU for sign off to complete the process.

### 6.8 LOSS TO FOLLOW-UP

Every effort should be made to follow-up patients who have been randomised. Patients should, if possible, remain under the care of an oncologist for the duration of the trial. If the care of a patient is returned to the General Practitioner, it is still the responsibility of the investigator to ensure that the follow-up data required by the protocol are collected and reported.

Patients will be asked to consent for follow up via national registries at registration.
7 SAFETY REPORTING

The principles of GCP require that both investigators and sponsors follow specific procedures when notifying and reporting adverse events or reactions in clinical trials. These procedures are described in this section. Section 7.3 - Investigator Responsibilities gives details of the investigator responsibilities and Section 7.4 - MRC CTU at UCL Responsibilities provides information on MRC CTU at UCL responsibilities.

7.1 DEFINITIONS

The definitions of the EU Directive 2001/20/EC Article 2 based on the principles of GCP apply to this comparison protocol. These definitions are given in Table 7.1: Definitions.

7.1.1 MEDICINAL PRODUCTS

An investigational medicinal product is defined as the tested investigational medicinal product and the comparators used in the trial (EU guidance ENTR/CT 3, April 2006 revision).

Adverse reactions include any untoward or unintended response to drugs. Reactions to AZD1775 should be reported appropriately.

7.1.2 ADVERSE EVENTS

Adverse Events include:

- An exacerbation of a pre-existing illness
- An increase in frequency or intensity of a pre-existing episodic event or condition
- A condition (even though it may have been present prior to the start of the trial) detected after trial drug administration
- Continuous persistent disease or a symptom present at baseline that worsens following administration of the trial treatment

7.1.3 EXEMPTED ADVERSE EVENTS

Adverse Events do not include:

- Medical or surgical procedures; the condition that leads to the procedure is the adverse event
- Pre-existing disease or a condition present before treatment that does not worsen
- Hospitalisations where no untoward or unintended response has occurred, e.g. elective cosmetic surgery, social admissions
- Overdose of trial medication without signs or symptoms
<table>
<thead>
<tr>
<th>SAFETY EVENT</th>
<th>DEFINITION</th>
</tr>
</thead>
<tbody>
<tr>
<td>Adverse Event (AE)</td>
<td>Any untoward medical occurrence in a patient or clinical trial subject to whom a medicinal product has been administered including occurrences that are not necessarily caused by or related to that product.</td>
</tr>
<tr>
<td>Adverse Reaction (AR)</td>
<td>Any untoward and unintended response to an investigational medicinal product related to any dose administered.</td>
</tr>
<tr>
<td>Unexpected Adverse Reaction (UAR)</td>
<td>An adverse reaction, the nature or severity of which is not consistent with the information about the medicinal product in question set out in the Summary of Product Characteristics (SPC) or Investigator Brochure (IB) for that product.</td>
</tr>
</tbody>
</table>
| Serious Adverse Event (SAE) or Serious Adverse Reaction (SAR) or Suspected Unexpected Serious Adverse Reaction (SUSAR) | Respectively any adverse event, adverse reaction or unexpected adverse reaction that:  
- Results in death  
- Is life-threatening*  
- Requires hospitalisation or prolongation of existing hospitalisation**  
- Results in persistent or significant disability or incapacity  
- Consists of a congenital anomaly or birth defect  
- Is another important medical condition*** |

*The term life-threatening in the definition of a serious event refers to an event in which the patient is at risk of death at the time of the event; it does not refer to an event that hypothetically might cause death if it were more severe, for example, a silent myocardial infarction.

**Hospitalisation is defined as an inpatient admission, regardless of length of stay, even if the hospitalisation is a precautionary measure for continued observation. Hospitalisations for a pre-existing condition (including elective procedures that have not worsened) do not constitute an SAE.

*** Medical judgement should be exercised in deciding whether an AE or AR is serious in other situations. The following should also be considered serious: important AEs or ARs that are not immediately life-threatening or do not result in death or hospitalisation but may jeopardise the subject or may require intervention to prevent one of the other outcomes listed in the definition above; for example, a secondary malignancy, an allergic bronchospasm requiring intensive emergency treatment, seizures or blood dyscrasias that do not result in hospitalisation or development of drug dependency.
7.1.4 DISEASE-RELATED EVENTS
Due to the seriousness of the disease in FOCUS4, the following situations that fulfil the definition of an SAE are excluded from expedited notification on an SAE CRF and should be reported on the Toxicity and Symptom, Progress or Death eCRFs as appropriate:

- Elective hospitalisation and surgery for treatment of metastatic colorectal cancer or its complication e.g. bowel obstruction
- Elective hospitalisation to simplify treatment or procedures
- Elective hospitalisation for pre-existing conditions that, in the investigator’s opinion have not been exacerbated by trial treatment
- Disease progression leading to hospitalisation or prolongation of hospitalisation or death as a result of disease progression.
- Death due to progressive CRC.

7.2 OTHER NOTABLE EVENTS

7.2.1 PREGNANCY
Pregnancy occurring during a patient’s participation in FOCUS4 should be reported on an SAE CRF and sent to the MRC CTU at UCL within 24 hours of the investigator becoming aware of the event. The outcome of the pregnancy should be carefully followed with updated information sent to the MRC CTU at UCL.

Any pregnancy that occurs in a female trial subject will be followed to termination or to term. Follow-up of a child born to a pregnant trial subject given active treatment will be as per standard clinical care. The clinical team responsible will be informed of the participation of the child’s mother in the trial and will be asked to inform the MRC CTU at UCL if there is any suspicion of any adverse effect of the trial medication.

Follow-up of a child born to the partner of a male trial subject given trial treatment will be as per standard clinical care. The clinical team responsible will be informed of the participation of the child’s father in the trial and will be asked to inform the MRC CTU at UCL if there is any suspicion of any adverse effect of the trial medication.

7.3 INVESTIGATOR RESPONSIBILITIES
Once a patient is randomised to FOCUS4-C, all non-serious AEs and ARs, whether expected or not, should be recorded in the patient’s medical notes and reported on the Toxicity and Symptom eCRF. SAEs and SARs should be notified to the MRC CTU at UCL within 24 hours of the investigator becoming aware of the event.
7.3.1 INVESTIGATOR ASSESSMENT

7.3.1.A Seriousness
When an AE or AR occurs, the investigator responsible for the care of the patient must first assess whether or not the event is serious using the definition given in Table 7.1. If the event is serious and not only related to disease progression, then an SAE CRF must be completed and the MRC CTU at UCL notified within 24 hours.

7.3.1.B Severity or Grading of Adverse Events
The severity of all AEs and/or ARs (serious and non-serious) in this trial should be graded using the NCI CTCAE v4.0. A version is summarised for FOCUS4-C in Appendix VII-C. However for a full list, please refer to http://evs.nci.nih.gov/ftp1/CTCAE/About.html.

Figure 7.1 is a flowchart at the end of this section to help explain the notification procedures. Any questions concerning this process should be directed to the MRC CTU at UCL in the first instance.

7.3.1.C Causality
The investigator must assess the causality of all serious events or reactions in relation to the trial therapy using the definitions in Table 7.2: Assigning Type of SAE Through Causality. There are five categories: unrelated, unlikely, possible, probable, and definitely related. If the causality assessment is unrelated or unlikely to be related, the event is classified as an SAE. If the causality is assessed as possible, probable or definitely related, then the event is classified as an SAR.
Table 7.2: Assigning Type of SAE Through Causality

<table>
<thead>
<tr>
<th>RELATIONSHIP</th>
<th>DESCRIPTION</th>
<th>SAE TYPE</th>
</tr>
</thead>
<tbody>
<tr>
<td>Unrelated</td>
<td>There is no evidence of any causal relationship.</td>
<td>Unrelated SAE</td>
</tr>
<tr>
<td>Unlikely</td>
<td>There is little evidence to suggest that there is a causal relationship (for example, the event did not occur within a reasonable time after administration of the trial medication). There is another reasonable explanation for the event (for example, the patient’s clinical condition, other concomitant treatment).</td>
<td>Unrelated SAE</td>
</tr>
<tr>
<td>Possible</td>
<td>There is some evidence to suggest a causal relationship (for example, because the event occurs within a reasonable time after administration of the trial medication). However, the influence of other factors may have contributed to the event (for example, the patient’s clinical condition and other concomitant treatments).</td>
<td>SAR</td>
</tr>
<tr>
<td>Probable</td>
<td>There is evidence to suggest a causal relationship and the influence of other factors is unlikely.</td>
<td>SAR</td>
</tr>
<tr>
<td>Definitely</td>
<td>There is clear evidence to suggest a causal relationship and other possible contributing factors can be ruled out.</td>
<td>SAR</td>
</tr>
</tbody>
</table>

If an SAE is considered to be related to trial treatment and drug is stopped or the dose modified, refer to Section 5.2.3 - Dose Modifications, Interruptions & Discontinuations.

7.3.1.D Expectedness
If there is at least a possible involvement of the trial treatment, the Investigator must make the initial assessment of the expectedness of the event; however the sponsor has the final responsibility for the determination of the expectedness. An unexpected adverse reaction is one not previously reported in the current Investigator Brochure (IB)/ Summary of Product Characteristics (SPC) or one that is more frequent or more severe than previously reported. The definition of an unexpected adverse reaction (UAR) is given in Table 7.1: Definitions. Please see Appendix VIII-C for a list of expected toxicities associated with the drug(s) being used in this trial. If a SAR is assessed as being unexpected, it becomes a SUSAR.

7.3.1.E Notification
The MRC CTU at UCL should be notified of all SAEs within 24 hours of the investigator becoming aware of the event.

Investigators should notify the MRC CTU at UCL of all SAEs occurring from the time of randomisation until 30 days after the last protocol treatment administration. SARs and SUSARs must be notified to
the MRC CTU at UCL until trial closure. Any subsequent events that may be attributed to treatment should be reported to the MHRA using the yellow card system.

7.3.2 NOTIFICATION PROCEDURE

1. The SAE CRF must be completed by the investigator (the consultant named on the Signature List and Delegation of Responsibilities Log who is responsible for the patient’s care or a co-investigator who has been delegated this responsibility by the responsible investigator), with due care being paid to the grading, causality and expectedness of the event as outlined above. In the absence of the responsible investigator or co-investigator, the form should be completed and signed by a member of the site trial team and faxed. The responsible investigator or co-investigator should subsequently check the SAE CRF, make changes as appropriate, sign and then re-fax to the MRC CTU at UCL as soon as possible. The initial report must be followed by detailed, written reports as appropriate.

   The minimum criteria required for reporting an SAE are the trial number and date of birth, name of investigator reporting and why the event is considered serious.

2. The SAE CRF must be sent by fax to the MRC CTU at UCL Fax: 020 7670 4818

3. Follow-up: patients must be followed up until clinical recovery is complete and laboratory results have returned to normal or baseline, or until the event has stabilised. Follow-up should continue after completion of protocol treatment if necessary. A further SAE CRF, indicated as ‘Follow-up’ should be completed and faxed to the MRC CTU at UCL as information becomes available. Extra, annotated information and/or copies of test results may be provided separately. The patient must be identified by trial number, date of birth and initials only. The patient’s name should not be used on any correspondence and should be deleted from any test results.

4. Staff should follow their institution’s procedure for local notification requirements.

SAE REPORTING

Within 24 hours of becoming aware of an SAE, please fax a completed SAE CRF to the MRC CTU at UCL on:
Fax: 020 7670 4818

7.4 MRC CTU at UCL RESPONSIBILITIES

Medically-qualified staff at the MRC CTU at UCL and/or the Chief Investigator (or a medically-qualified delegate) will review all SAE reports received. The causality assessment given by the local investigator at the hospital cannot be overruled; in the case of disagreement, both opinions will be provided in any subsequent reports.
The MRC CTU at UCL is undertaking the duties of Trial Sponsor and is responsible for the reporting of SUSARs and other SARs to the regulatory authorities (MHRA) and the research ethics committees, as appropriate. Fatal and life-threatening SUSARs must be reported to the competent authorities within 7 days of the MRC CTU at UCL becoming aware of the event; other SUSARs must be reported within 15 days.

AstraZeneca will provide updated safety information and the MRC CTU at UCL will also keep all investigators informed of any safety issues that arise during the course of the trial.

The MRC CTU, as Sponsor, will submit Development Safety Update Reports (DSUR) to Competent Authorities (Regulatory Authorities) and Ethics Committees.

AstraZeneca will be notified of all reportable (serious and unexpected and drug-related/unknown relationship) events and the MRC CTU at UCL will also provide AstraZeneca with a copy of the DSUR.

**Figure 7.1: Safety Reporting Flow Chart**

- Adverse Event/Adverse Reaction
  - Was the event serious?
    - Resulted in death
    - Life threatening
    - Required inpatient hospitalisation or prolongation of existing hospitalisation
    - Persistent or significant disability/incapacity
    - Congenital anomaly/birth defect
  - No
  - Yes
    - Was the SAE specified in the protocol as being exempt from expedited reporting?
      - No
      - Record on an SAE CRF and notify the MRC CTU within 24 hours of becoming aware of the event
      - Yes
        - Yes
          - AE
            - Record on the Toxicity and Symptom, Progress or Death CRFs as appropriate
        - No
          - SAE
            - Record on the Toxicity and Symptom, Progress or Death CRF as appropriate
8 QUALITY ASSURANCE & CONTROL

8.1 RISK ASSESSMENT

The Quality Assurance (QA) and Quality Control (QC) considerations will be based on a formal Risk Assessment, which will acknowledge the risks associated with the conduct of the trial and how to address them with QA and QC processes. QA includes all the planned and systematic actions established to ensure the trial is performed and data generated, documented and/or recorded and reported in compliance with the principles of GCP and applicable regulatory requirements. QC includes the operational techniques and activities done within the QA system to verify that the requirements for quality of the trial-related activities are fulfilled. This Risk Assessment will be reviewed by the Research Governance Committee (RGC) within the MRC CTU at UCL and this will be used to develop the Data Management, Safety and Monitoring Plans, which will be kept separately.

8.2 CENTRAL MONITORING AT MRC CTU at UCL

MRC CTU at UCL staff will perform checks on CRFs data for errors, inconsistencies and missing data points.

Other essential trial issues, events and outputs will be detailed in the Data Management, Safety and Monitoring Plans that is based on the trial-specific Risk Assessment.

8.3 ON-SITE MONITORING

The frequency, type and intensity for routine monitoring and the requirements for triggered monitoring will be detailed in the Data Management, Safety and Monitoring Plans. This plan will also detail the procedures for review and sign-off.

8.3.1 DIRECT ACCESS TO PATIENT RECORDS

Participating investigators should agree to allow trial-related monitoring, including audits, ethics committee review and regulatory inspections by providing direct access to source data and documents as required. Patients’ consent for this must be obtained.

8.3.2 CONFIDENTIALITY

The principles of the UK DPA will be followed.
9 STATISTICAL CONSIDERATIONS

9.1 METHOD OF RANDOMISATION

This is described fully in the Master Protocol. Patients in FOCUS4-C will be allocated to their trial treatment using minimisation with a random element stratified by various factors known or thought to be prognostic of outcome as well as the type of first-line treatment regime.

9.2 OUTCOME MEASURES

The primary outcome measure for FOCUS4-C is progression-free survival (PFS) timed from randomisation at the end of first-line treatment. PFS includes deaths from any cause or progression of disease (PD according to RECIST v1.1 criteria) based upon CT scan. Secondary outcome measures include safety, toxicity and Objective Response Rate (ORR).

9.3 SAMPLE SIZE

All the molecular cohort comparisons within FOCUS4 are adaptive in design. The generic sample size calculations are provided in the Master Protocol but these have been modified for the agent being tested in this trial. See Tables 9.1, 9.2 and 9.3 for the expected prevalence and operating characteristics.

9.4 INTERIM MONITORING & ANALYSES

See the Master protocol for more detail on the role of the IDMC. Safety data from the tolerability phase will be reviewed by the IDMC when 12 patients randomised to AZD1775 have completed 2 cycles of treatment (see section 5.2.3 for details). Data from interim analysis at Stage I will be reviewed by the IDMC and results will be released to allow an open decision to be made about whether to proceed to Stage II. Lack of sufficient activity stopping guidelines are inbuilt into the study design. Early stopping for efficacy is unlikely and the IDMC would make this recommendation only if the result is likely to convince a broad range of clinicians including those entering patients into the trial and the clinical community. Anticipated timelines for the stages are provided in Tables 9.2 and 9.3. An IDMC Charter has been drawn up that describes the membership of the IDMC, relationships with other committees, terms of reference, decision-making processes, and the timing and frequency of interim analyses.
9.5 ANALYSIS PLAN (BRIEF)

Please see the FOCUS4 Master Protocol Section 9.5.

Biomarker-drug combinations to be tested:

<table>
<thead>
<tr>
<th>Biomarker</th>
<th>Expected prevalence</th>
<th>Drug</th>
</tr>
</thead>
<tbody>
<tr>
<td>H3K36me3 loss</td>
<td>6%</td>
<td>WEE1 inhibitor (AZD1775)</td>
</tr>
<tr>
<td>p53 mutant + RAS mutant</td>
<td>23%</td>
<td>WEE1 inhibitor (AZD1775)</td>
</tr>
</tbody>
</table>

We propose a 2-stage design derived from the MRC CTU Multi-Arm Multi-Stage (MAMS) design, randomising patients to active drug vs Active Monitoring in a 2:1 ratio with an early assessment of Objective Response Rate (ORR) after 16 weeks of drug in the active arm. In addition, a Stage I interim analysis exploring progression-free-survival (PFS) in the maintenance setting would be completed.

Summary of operating characteristics for each molecular comparison:

Each molecular comparison has been powered separately. See Tables 9.2 and 9.3 for design characteristics. Based upon the prevalence of each biomarker, we have assumed a recruitment rate of 2 and 7 patients per month into the H3K36me3 loss and the p53/RAS mutant cohorts respectively. We assume a median PFS in the control arm of 3.6 months and seek a target HR of 0.5 for the less biologically enriched p53/RAS mutant cohort and a HR of 0.4 in the more biologically enriched H3K36me3 loss cohort. Power is kept high for the stage I analysis (95%) and then maintained at 80% overall.

Operating characteristics for the H3K36me3 loss group (6%, 2 per month)

<table>
<thead>
<tr>
<th>TABLE 9.2</th>
<th>Stage I Safety and LSA</th>
<th>Stage II Efficacy</th>
</tr>
</thead>
<tbody>
<tr>
<td>Target HR for PFS</td>
<td>0.4</td>
<td>0.4</td>
</tr>
<tr>
<td>1-sided alpha</td>
<td>0.4</td>
<td>0.025</td>
</tr>
<tr>
<td>Power (overall power maintained at 80%)</td>
<td>95%</td>
<td>81%</td>
</tr>
<tr>
<td>Critical HR</td>
<td>0.90</td>
<td>0.54</td>
</tr>
<tr>
<td>Time required (months)</td>
<td>17</td>
<td>11</td>
</tr>
<tr>
<td>Cumulative time (months)</td>
<td>17</td>
<td>28</td>
</tr>
<tr>
<td>Cumulative events required in control arm (total)</td>
<td>8 (19)</td>
<td>15 (37)</td>
</tr>
<tr>
<td>Total expected cumulative randomisations (no. in active arm)</td>
<td>34 (23)</td>
<td>55 (37)</td>
</tr>
<tr>
<td>Total expected number with ORR outcome in active arm</td>
<td>18</td>
<td>32</td>
</tr>
</tbody>
</table>

*LSA= Lack-of-sufficient-activity*
Operating characteristics for the P53 mutant and RAS mutant group (23%, 7 per month)

<table>
<thead>
<tr>
<th>TABLE 9.3</th>
<th>ORR early activity</th>
<th>Stage I Safety and LSA</th>
<th>Stage II Efficacy</th>
</tr>
</thead>
<tbody>
<tr>
<td>Target HR for PFS</td>
<td>-</td>
<td>0.5</td>
<td>0.5</td>
</tr>
<tr>
<td>1-sided alpha</td>
<td>-</td>
<td>0.4</td>
<td>0.025</td>
</tr>
<tr>
<td>Power (overall power maintained at 80%)</td>
<td>-</td>
<td>95%</td>
<td>81%</td>
</tr>
<tr>
<td>Critical HR</td>
<td>-</td>
<td>0.92</td>
<td>0.63</td>
</tr>
<tr>
<td>Time required (months)</td>
<td>9</td>
<td>10</td>
<td>6</td>
</tr>
<tr>
<td>Cumulative time (months)</td>
<td>9</td>
<td>10</td>
<td>16</td>
</tr>
<tr>
<td>Cumulative events required in control arm (total)</td>
<td>-</td>
<td>13 (30)</td>
<td>26 (63)</td>
</tr>
<tr>
<td>Total expected cumulative randomisations (no. in active arm)</td>
<td>-</td>
<td>70 (47)</td>
<td>113 (75)</td>
</tr>
<tr>
<td>Total expected number with ORR outcome in active arm</td>
<td>25</td>
<td>33</td>
<td>61</td>
</tr>
</tbody>
</table>

* LSA= Lack-of-sufficient-activity
10 ANCILLARY STUDIES

Ancillary studies for FOCUS4-C are broadly described in the Master Protocol; a specific focus will be identification of other DNA repair biomarkers which may further identify patients most likely to respond to WEE1 inhibition.

Please also refer to the Sample Collection and Handling SOP.
11 OTHER ISSUES

The following sections are not documented in this protocol as they are common to all comparisons in the FOCUS4 Trial Programme. Please refer to the FOCUS4 Master Protocol for:

- Regulatory & Ethical issues (Section 11)
- Indemnity (Section 12)
- Finance (Section 13)
- Oversight & Trial Committees (Section 14)
- Publications (Section 15)
12 PROTOCOL AMENDMENTS

Please check on the FOCUS4 website (www.focus4trial.org) or with the MRC CTU FOCUS4 Trial Managers to confirm the most recent version of the FOCUS4 protocols and associated documents. This is the second signed agreed version of the FOCUS4-C Protocol and the first version of the FOCUS4-C appendices.

Please note this section will only refer to the FOCUS4-C protocol and its associated documents. Each separate comparison protocol will document all protocol amendments separately.

1) FOCUS4-C Protocol version 1.0 to version 2.0

A summary FOCUS4-C comparison protocol section V1.0 (01-Feb-2013) outlining the intention for this comparison was included as part of the FOCUS4 Trial Programme protocol and was approved as part of the original submission. Since V1.0 of this document, the choice of interventional agent and molecular cohort suitable for FOCUS4-C has changed. Version 1 included an outdated trial schema. Originally this biomarker cohort had just been the RAS mutant group. An initial discussion had been to include these drugs but the combination did not show promising results. Therefore an alternative agent was progressed for a RAS/p53 mutant group. We will no longer be testing a combined AKT and MEK inhibitor in patients with a KRAS or NRAS mutations, but will be testing Wee1 inhibitor (AZD1775) instead in patients with H3K36me3 loss or combined p53 and RAS mutations. As a result, the FOCUS4-C section of the protocol has been completely re-written.

2) FOCUS4-C Protocol Version 2.0 to Version 3.0 and FOCUS4-C Appendices Version 1.0 to Version 2.0

FOCUS4-C protocol V2.0 and FOCUS4-C appendices V1.0 were updated following the Letter of Non-acceptance of substantial amendment 7 from MHRA to address their comments and to correct minor errors.

- Updates to assessment schedule to specify that pulse rate, blood pressure and weight measurements should be performed as part of the clinical evaluation.
- Addition of pregnancy testing every 3 weeks for women of child bearing potential randomised to the AZD1775 arm.
- Clarification that adequate contraception should be used for two weeks prior to starting trial treatment and for one month following end of treatment for women of child bearing potential.
- Addition of guidance regarding counselling of male participants wishing to father children while taking AZD1775 and three months after stopping treatment regarding freezing of sperm samples.
- Addition of exclusion criteria to exclude patients at risk of GI haemorrhage:
  - Active or previous peptic ulceration unless well-controlled on PPIs for at least 6 months or resolved with HP eradication.
  - Previous gastrointestinal bleeding except where the cause of the bleeding has been surgically removed.
• Addition of guidance to the protocol and appendices to state that NSAIDs should be used with caution with AZD1775 to the protocol and appendices.

3) FOCUS4-C Protocol Version 3.0 to Version 4.0 and FOCUS4-C Appendices Version 3.0 to Version 4.0
• A change to the dosing schedule has been made and all relevant sections have been updated, including changes to the PIS and diary cards.
• A tolerability phase restricted to the Level 3 sites has been included for the first 12 patients treated on AZD1775.
• A new IB has been approved and necessary changes have been made to the background section, dose modification section and expected toxicities.
• Addition of ECG assessment prior to each cycle for patients receiving AZD1775
• Some minor changes to the CTU staff contact details have been updated.

4) FOCUS4-C Protocol Version 4.0 to Version 5.0 and FOCUS4-C Appendices Version 4.0 to Version 5.0
• Removal of the biomarker hierarchy that previously determined that a patient with multiple mutations could only be offered enrolment into one molecular comparison (the exception being the BRAF mutation).
• Update of MRC CTU at UCL office address.
• Some minor changes to MRC CTU staff contact details have been updated.
• Minor corrections to spelling and grammar.
• Clarification around the washout period when stopping treatment with AZD1775.
• Clarification on the ECG assessment schedule at the start of treatment.
13 REFERENCES


