CTEP # 9350
Clinical Center # 13-C-0032

TITLE: A Phase I Study of Single-agent AZD1775 (MK-1775), a Wee1 Inhibitor, in Patients with Advanced Refractory Solid Tumors

Abbreviated Title: Ph I study of AZD1775

Coordinating Center:	Developmental Therapeutics Clinic (NCIDTC) National Cancer Institute 10 Center Drive Bethesda, MD 20892
Principal Investigator:	Naoko Takebe, MD, PhD[A-E] 31 Center Drive Bldg 31 Room 3A44 Bethesda, MD 20892 Phone: (240) 781-3398 Fax: (240) 541-4515 takeben@mail.nih.gov
NCI Associate Investigators:	A. P. Chen, MD, DCTD[A-E] James H. Doroshow, MD, DCTD[A-E] Richard Piekarz, MD, PhD, DCTD[A-E] Howard Streicher, MD CTEP, DCTD[A-E] Geraldine O'Sullivan Coyne, MD, PhD, CCR[A-E] Sheila Prindiville, MD, MPH, DCTD[A-E]
Non-NCI Associate Investigators:	Lamin Juwara, CRNP Leidos Biomed, FNLCR[A,B,E]
Referral Contact:	Ashley Bruns, RN[A,B,E] Bldg 10/Rm 8D53 Phone: (240) 858-3162; Fax: (301) 451-5625 ashley.bruns@nih.gov
Statistician:	Larry Rubinstein, PhD, BRB/DCTD[E] 9609 Medical Center Dr. Rm 5W106 Phone: (240) 276-6026 rubinsteinl@mail.nih.gov

NCI Supplied Agent:	AZD1775 (MK-1775) (NSC 751084)
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PRÉCIS

Background:

- Wee1 is a tyrosine kinase involved in the phosphorylation and inactivation of cyclin-dependent kinase 1 (CDK1/CDC2)-bound cyclin B, resulting in G2 cell cycle arrest in response to DNA damage to allow time for DNA repair. Recent preclinical data additionally implicates Wee1 in maintenance of genomic integrity during S phase.
- AZD1775 (formerly known as MK-1775) is a selective inhibitor of Wee1 kinase. Recent preclinical model data additionally show single agent anti-tumor activity in multiple cancer cell lines and tumor xenografts.
- Preliminary data show AZD1775 is tolerable at lower doses in combination with chemotherapeutic agents. We propose to demonstrate single-agent activity for AZD1775.

Primary Objective:

- To establish the safety and tolerability of single-agent AZD1775 in patients with refractory solid tumors
- To determine the pharmacokinetics of AZD1775 in patients with refractory solid tumors

Secondary Objectives:

- To evaluate the antitumor activity of AZD1775 in patients with refractory solid tumors

Exploratory Objectives:

- To determine the effect of AZD1775 on markers of DNA damage and apoptosis in tumor tissue and circulating tumor cells
- To assess whether sufficient Wee1 inhibition is maintained throughout the therapeutic regimen
- To identify tumor genomic alterations and gene expression patterns potentially associated with AZD1775 antitumor activity

Eligibility:

- Patients must have histologically confirmed solid tumors for which all standard therapy known to prolong survival have failed, or for which standard therapies do not exist.
- No major surgery, radiation, or chemotherapy within 3 weeks (or 5 half-lives, whichever is shorter) prior to entering the study
- Adequate organ function

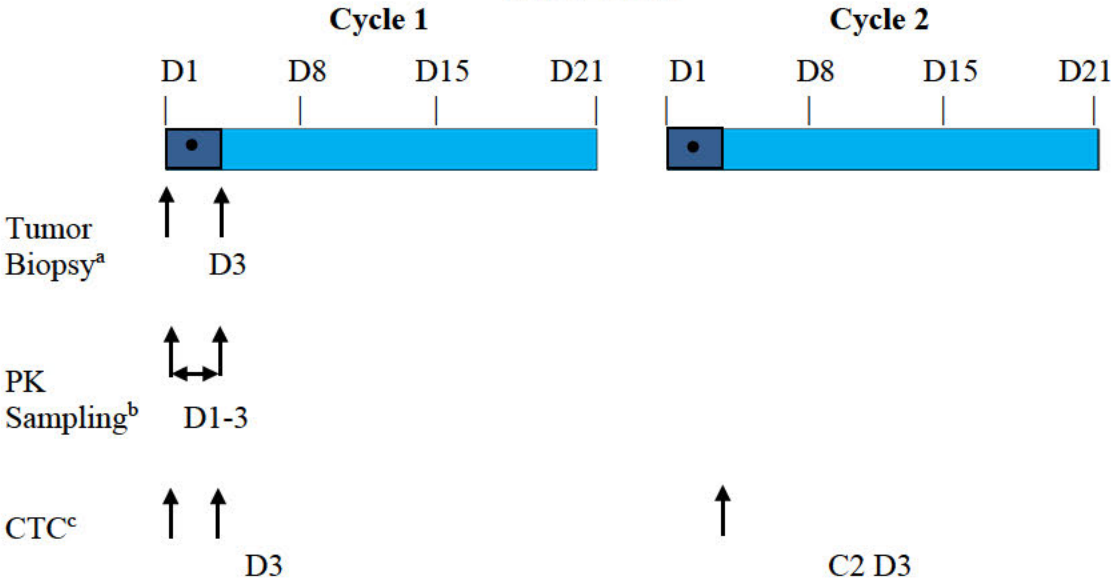
Study Design:

- This study will follow a traditional 3+3 design.
- In Arm A starting at dose level 1, AZD1775 will be administered orally, BID, for 5 doses (D1-3) during each cycle. Starting at dose level 2 and onwards, AZD1775 will be administered orally, BID, for 5 doses for the first 2 weeks of each cycle (D1-3 and 8-10). Each cycle is 21 days (± 1 day for scheduling).
- Once MTD is established, 6 additional patients will be enrolled at the MTD to further evaluate that dose for PK and PD endpoints.
- A further expansion arm of 6 additional patients with documented tumors harboring BRCA-1 or -2 mutations will also be enrolled at the MTD to further explore the safety of the agent and obtain preliminary evidence of activity in this patient population.

- Based on preliminary evidence of drug activity in an alternative once-daily dosing schedule, patients without a documented BRCA mutation will be accrued to a once-daily dosing schedule Arm B, with mandatory paired tumor biopsies at the maximum tolerated single daily dose, to further evaluate PD endpoints. AZD1775 will be administered orally once daily for 5 days (D1-5 and 8-12) during weeks 1 and 2 of each 21-day cycle (± 1 day for scheduling).
- During the escalation phase, tumor biopsies will be optional and will be evaluated for pharmacodynamic (PD) studies for evidence of Wee1 inhibition, DNA damage and repair, and apoptosis (γH2AX, pNbs1, Rad51, pTyr15-Cdk, and caspase 3). During the expansion phase, once MTD is reached, mandatory paired tumor biopsies will be pursued in up to 20 additional patients enrolled at the MTD to further evaluate PD endpoints.

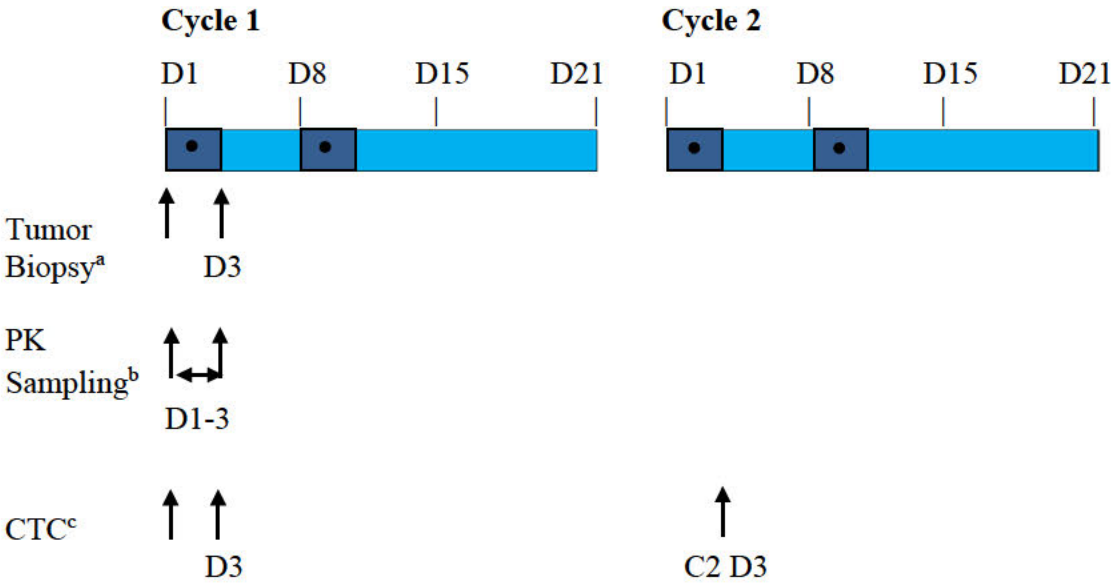
SCHEMA (Arm A)

Dose Level 1*



* For DL1/DL-1 only, AZD1775 will be given orally for a total of 5 doses the first week of each cycle.

Dose Level 2 and Higher*



* For DL2 and higher, AZD1775 will be given orally for a total of 5 doses during the first and second week of each cycle.

a During the escalation phase, optional paired tumor biopsies will be performed at baseline (pre-treatment) and 2-5 hours after the 5th dose of AZD1775 on C1D3. In instances where there is a scheduling conflict, biopsies may be performed within $\pm$ 1 day of the 5th dose of C1D3. During the expansion phase, paired tumor biopsies will be mandatory in the six additional patients enrolled at the MTD. Tumor biopsies will be optional in the 6-patient BRCA mutation MTD expansion arm.

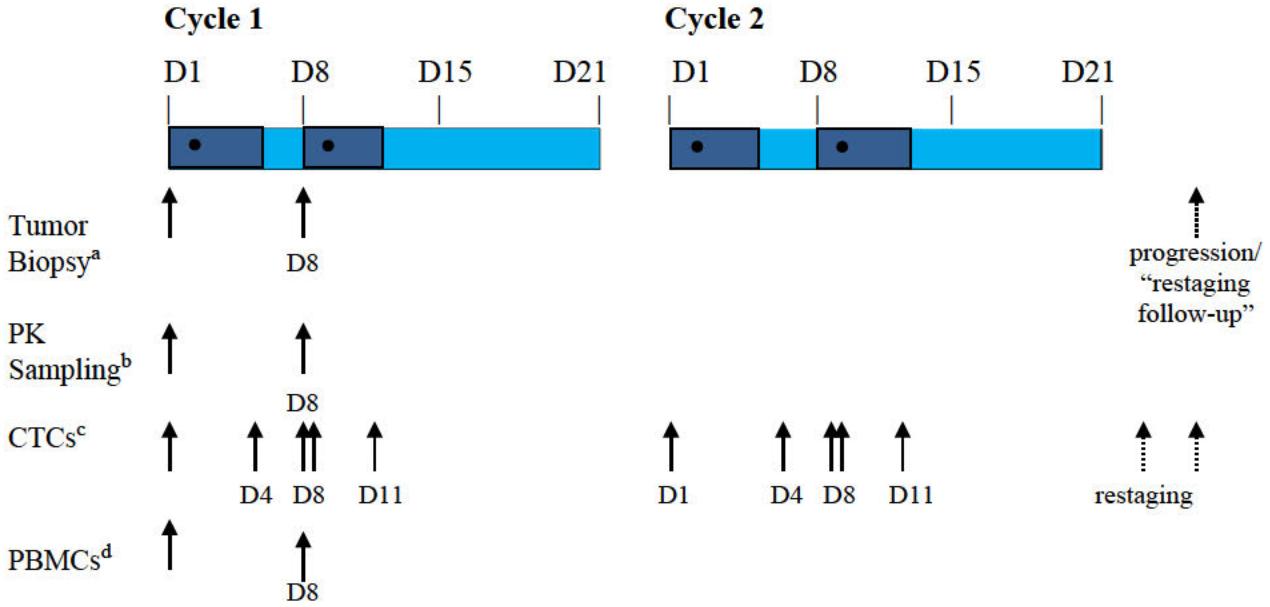
b Blood samples for PK analyses will be collected at the following timepoints:
C1D1: Prior to the initial first dose, and at 1, 2, 4, 6, and 8 hours after the first dose
C1D2: Prior to the 3rd dose administered on day 2
C1D3: Prior to the 5th dose, and 1, 2, 4, 6, and 8 hours after the 5th dose administered on day 3
Additional PK sampling will be optional (but encouraged). This sampling would occur 24 and 48 hours after the 5th dose of AZD1775, and will occur on Days 4 and 5, respectively (not reflected in Flow Chart).

c Circulating tumor cells will be obtained at baseline, and 2-5 hours after the 5th dose of AZD1775 during cycle 1 and cycle 2

Dose Escalation Schedule for Arm A	
Dose Level	***AZD1775 (orally BID) x 5 doses/weeks***
-1	200 mg BID x 5 doses (days 1-3) for 1 week
1	225 mg BID x 5 doses (days 1-3) for 1 week
2	225 mg BID x 5 doses (days 1-3 each week) for 2 weeks
3	300 mg BID x 5 doses (days 1-3 each week) for 2 weeks
4	400 mg BID x 5 doses (days 1-3 each week) for 2 weeks
5	550 mg BID x 5 doses (days 1-3 each week) for 2 weeks

With Amendment G (07/17/2014), patients without a documented BRCA mutation will be accrued to an alternative dosing schedule as described below (Arm B), based on new preclinical data demonstrating drug activity with once daily dosing. At the maximum tolerated single daily dose, mandatory paired tumor biopsies will be performed at baseline (pre-treatment) and, with Amendment M (11/20/16), prior to the 6th dose of AZD1775 on C1D8. Collection times for PD and PK samples are described below.

SCHEMA (Arm B)



a For Arm B, tumor biopsies will be optional during the escalation phase and mandatory during the expansion phase at the maximum tolerated dose; paired tumor biopsies will be performed at baseline (pre-treatment) and prior to the 6th dose of AZD1775 on C1D8. In instances where there is a scheduling conflict, biopsies may be performed within +/- 1 day of the 6th dose on C1D8. In addition, an optional "restaging follow-up biopsy" may be performed on day 1 (± 2 days) of the cycle following any restaging at which a 10-20% increase in tumor volume is observed (according to RECIST criteria) if the patient has been on study for at least 4 cycles, or at time of disease progression. (For example, a restaging follow-up biopsy would be performed on cycle 6 day 1 for a 10-20% tumor volume increase detected at the restaging following cycle 4).

b Blood samples for PK analyses will be collected at the following time points:
C1D1: Prior to the first dose and at 2 and 4 hr after the first dose,
C1D8: Prior to the 6th dose

c Circulating tumor cells (optional) will be obtained at the following time points during each cycle: on day 1 (prior to AZD1775 administration), on day 4 (4 ± 1 hours after AZD1775 administration), on day 8 prior to the dose of AZD1775, on day 8 (4 ± 1 hours after AZD1775 administration), and on day 11 (4 ± 1 hours after AZD1775 administration). Additional optional CTC samples will be obtained at each restaging and at time of disease progression or "restaging follow-up" biopsy (see above).

d Optional PBMC immune blood samples and serum will be collected at baseline and on day 8 prior to dosing (+/- 1 day) from Arm B patients.

Dose Escalation Schedule for Arm B	
Dose Level	***AZD1775 (orally once daily)***
-1	200 mg daily D1-3 each week for 2 weeks
1	200 mg daily D1-5 each week for 2 weeks
2	225 mg daily D1-5 each week for 2 weeks
3	250 mg daily D1-5 each week for 2 weeks
4	300 mg daily D1-5 each week for 2 weeks
5	400 mg daily D1-5 each week for 2 weeks
6	500 mg daily D1-5 each week for 2 weeks

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1 OBJECTIVES

1.1 Primary Objectives

- To establish the safety and tolerability of single-agent AZD1775 (formerly known as MK-1775) in patients with refractory solid tumors.
- To determine the pharmacokinetics of single-agent AZD1775 in patients with refractory solid tumors.

1.2 Secondary Objective

- To evaluate the antitumor activity of AZD1775 in patients with refractory solid tumors.

1.3 Exploratory Objectives

- To determine the effect of AZD1775 on markers of DNA damage and apoptosis in tumor tissue and circulating tumor cells (CTCs).
- To assess whether sufficient Wee1 inhibition is maintained throughout the therapeutic regimen.
- To identify tumor genomic alterations and gene expression patterns potentially associated with AZD1775 antitumor activity.

2 BACKGROUND

2.1 Cell Cycle Regulation

Cell cycle regulation depends on a delicate balance between cyclins, cyclin-dependent kinases (Cdks) that allow for progression through the cell cycle, and Cdk inhibitors that halt progression in the setting of DNA damage. DNA replication during S phase generates intermediate DNA structures that are vulnerable to genotoxic insults. In addition, oncogenes can induce lesions at replication forks, further exacerbating genomic instability. In response to DNA damage, surveillance protein complexes such as Mre11/Rad50-Nbs1 complex and Rad9/Rad1/Hus1 complex recruit repair proteins and halts cell cycle progression, allowing time for DNA repair. Ataxia-telangectasia mutated (ATM) protein kinase and ataxia-telangectasia-related (ATR) protein kinase, two members of the phosphatidyl inositol 3-kinase-like kinase (PIKK) family, are central to the DNA damage repair process. Depending on the type of genotoxic stress, either ATM or ATR is preferentially activated[1]. ATM is primarily activated in response to double-stranded DNA breaks, while ATR is activated by a broader spectrum of genotoxic stimuli and primarily activated in response to single-strand DNA breaks.

ATM/ATR kinases regulates cell cycle checkpoints by phosphorylation of multiple downstream proteins, including proteins associated with the recognition of double-stranded DNA breaks such histone H2AX, proteins involved in the assembly of protein complexes at the site of DNA damage, and effector kinases such as checkpoint kinase 1/2

(Chk1) and (Chk2). In contrast to Chk2, Chk1 can be activated by both ATM and ATR, and activation can occur during normal cell cycle progression or in response to replicative stress[2]. Activated Chk1 in turn concomitantly phosphorylates Wee1 and Cdc25C, thereby activating Wee1 kinase activity and inactivating Cdc25C phosphatase activity. Wee1 is a tyrosine kinase implicated in the inhibitory phosphorylation of CDK1/CDC2-bound cyclin B complex responsible for G2 arrest[3]. Originally identified in fission yeast[4], wee 1 deficiency resulted in premature mitotic entry and replication of smaller-sized yeast. Wee1 belongs to a family of protein kinases involved in the terminal phosphorylation and inactivation of (CDK1/CDC2)-bound cyclin B via phosphorylation of its Tyr15 residue near the ATP-binding pocket (pTyr15-Cdk), resulting in G2 cell cycle arrest in response to DNA damage. Wee1 overexpression has been demonstrated in hepatocellular carcinoma[5], luminal and HER-2 positive breast cancers[6], colon, lung carcinoma, and seminoma tumor samples[7].

2.2 AZD1775

Mechanism of Action

AZD1775, is a selective, adenosine-triphosphate (ATP) competitive, small molecule inhibitor of Wee1 kinase (IC_{50} = 5.18nM), that directly inhibits phosphorylation of CDC2 at Tyr15. A comprehensive review of AZD1775 can be found in the AZD1775 Investigator's Brochure (2017).

AZD1775 has been shown to inhibit phosphorylated CDC2 in p53 mutant WiDr colon and lung carcinoma cells, as measured by colorimetric ELISA in a dose-dependent manner [Figure 1A]. Reduction of pY15-Cdk levels was recently demonstrated in the A673 Ewing sarcoma xenograft model (included as Figure 7). Additionally, premature mitotic entry was demonstrated by concomitant increase in the percentage of cells expressing phosphorylated histone H3 at the same doses [Figure 1 B]. AZD1775 has demonstrated activity in multiple cancers based on in vitro activity in NCI 60 cell lines [Appendix B].

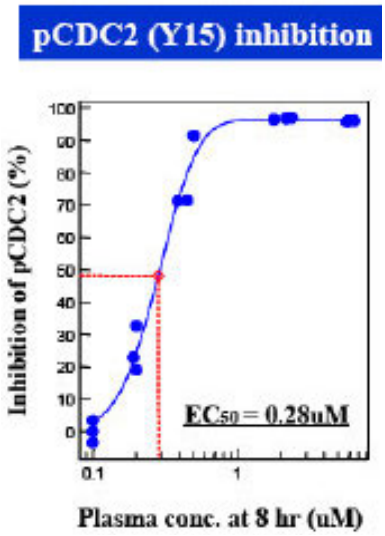


Figure 1A

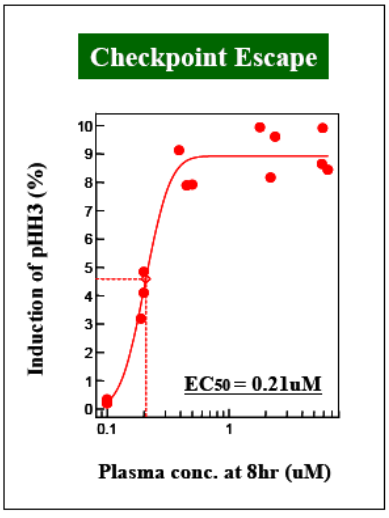


Figure 1B

2.3 Rationale

p53, a key regulator of the G1-S transition in cell cycle regulation, is frequently mutated in malignancies. The G1 checkpoint halts the progression of the cell cycle at the G1 phase, allowing time for repair of DNA damage and maintenance of genomic integrity before replication. As a result, many tumors have defective G1 checkpoint mechanisms and are dependent on an intact S-G2 checkpoint for progression through the cell cycle and continued growth. Early in vitro studies of ovarian, colon, cervical, and lung cancer cell lines exposed to Wee1 inhibition demonstrated radiation-induced G2 abrogation, with enhanced effect more notable in p53-deficient cell lines[8]. Additional support for correlation with p53 status was shown by Hirai and colleagues, where ovarian carcinoma cells expressing short hairpin RNA against p53 were more sensitive to chemotherapeutic agents than their wild-type counterparts[9].

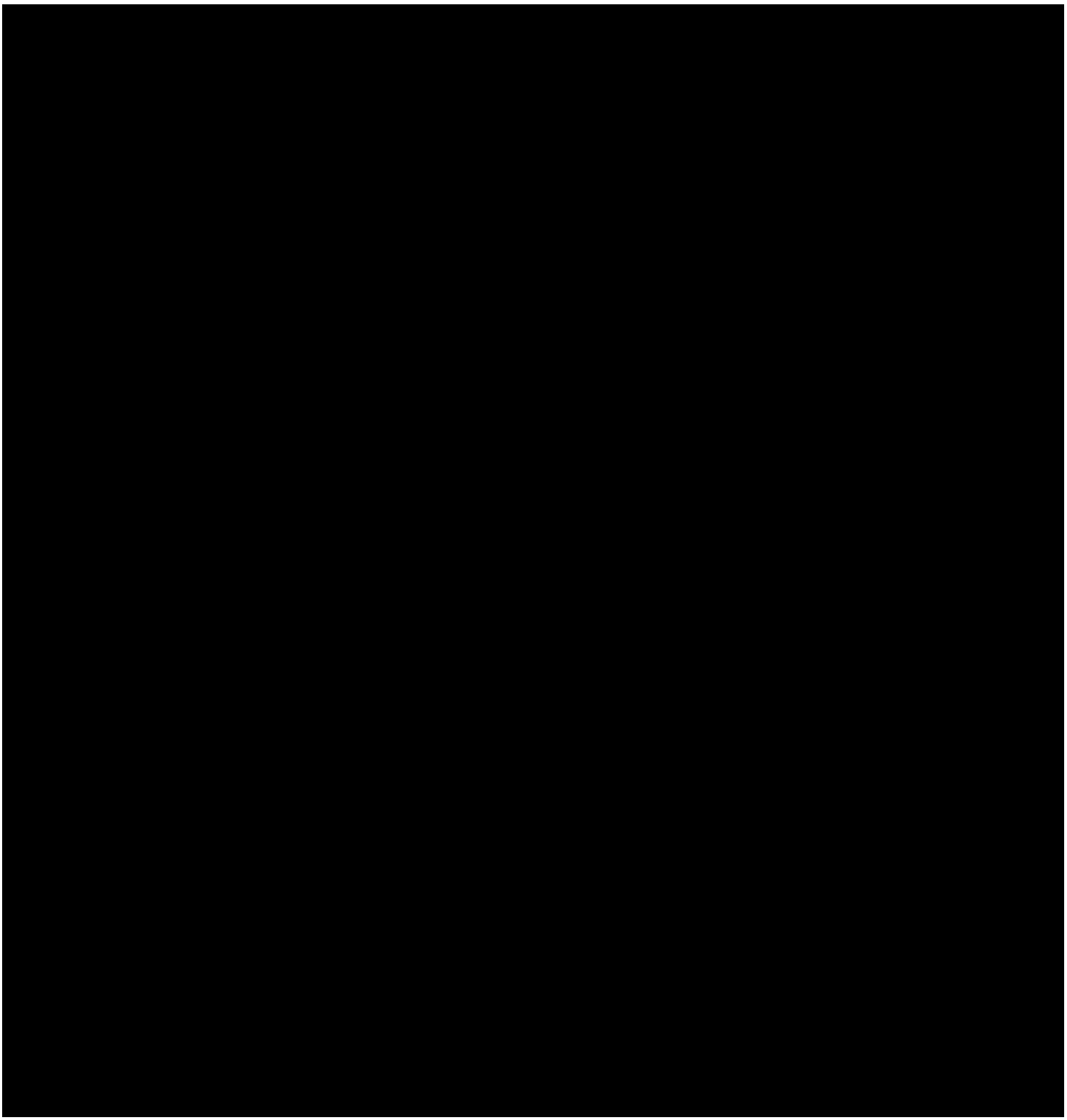
Preclinical Studies

Early preclinical data demonstrated enhancement of chemotherapeutic effect in combination with carboplatin, cisplatin, and gemcitabine in p53 deficient colon and lung carcinoma cells[9], and with 5-FU in p53-deficient colon and pancreatic cells, but not p53 wild-type colon cancer cells[10]. The same group also demonstrated objective tumor regression in xenograft models using immunodeficient nude rats subcutaneously bearing p53 mutant colon carcinoma exposed to AZD1775 in combination with gemcitabine, in xenografts bearing breast carcinomas exposed to combination with capecitabine[9], and in p53-deficient primary pancreatic cancer xenografts, in combination with gemcitabine[11].

[redacted]

Previous studies targeting Wee1 inhibition have focused on abrogation of the G2 checkpoint and mitotic lethality as a mechanism for enhancement of chemotherapeutic effect in combination with various DNA damaging agents. In addition to its role in the G2 checkpoint, recent data additionally implicates Wee1 in maintenance of genomic integrity during S phase[12,13]. In vitro, Wee1 depletion by siRNA rapidly induces DNA damage during S phase, accompanied by marked accumulation of ssDNA in replicating areas[14]. More recent in vitro studies performed by the same group, showed that Wee1 inhibition resulted in uncontrolled Cdk activity that forced unscheduled initiation of DNA replication, destabilization of replication forks, and massive induction of DNA breaks.

Wee1 has additionally been shown to modulate the activity of the Mus81-Eme1 endonuclease complex responsible for cleavage of stalled replication forks by introduction of double stranded breaks to allow for replication recovery[13]. Massive induction of DNA breaks eventually overwhelms cellular capacity to repair the lesions, and as Wee1 inhibition also results in abrogation of the G2 checkpoint, cells are pushed towards mitotic lethality secondary to inability to repair damaged DNA. Based on these recent data demonstrating the role of Wee1 in maintenance of genomic integrity, we propose that AZD1775 may have single agent activity.



[redacted]

Non-Clinical Pharmacokinetics

[redacted]

Metabolism

[redacted]

Non-clinical toxicology

[redacted]

[redacted]

Clinical Experience

[redacted]

At the time of Amendment G, dated 07/17/2014, 18 patients have been enrolled onto Arm A of this study and the MTD established at 225 mg twice daily for 5 doses each week, for two consecutive weeks[16]. Confirmed partial responses were observed in two patients carrying BRCA mutations: one with head and neck cancer and one with ovarian cancer. As of Amendment I (dated 8/6/15), a total of 10 patients have been accrued to Arm B (qd dosing); this schedule has been well tolerated to date but persistent grade 2 nausea and vomiting on DL2 necessitated expansion to 6 patients to better define the toxicity. At the time of Amendment M (dated 11/20/16), a total of 24 patients have been accrued to Arm B, and we have established the Arm B MTD as DL4 (300 mg once daily on days 1-5 each week for two weeks).

Clinical Efficacy

Of 176 evaluable patients who received AZD1775 (either single or multiple doses) as monotherapy or in combination with gemcitabine, cisplatin, or carboplatin on Study PN001, 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[17]. A phase II trial of AZD1775 in combination with carboplatin in patients with refractory or resistant TP53-mutated ovarian cancer (Study PN009) has also been performed, yielding a 43% overall response rate (partial response + complete response)[18].

Safety

[redacted]

[redacted]

In study PN009[18] and the pharmacokinetic sub-study to PN001[17] which used the pre-market formulation, (both combining 2.5-day BID dosing of AZD1775 with carboplatin) toxicities were not qualitatively different from the ones observed in the carboplatin arm of PN001. However, increased hematological toxicity was observed, which was attributed to a drug-drug interaction with aprepitant (which was administered as anti-nausea medication in these studies). Preliminary pharmacokinetic analyses revealed that co-administration of AZD1775 and aprepitant resulted in a ~40% increase of AZD1775 exposure.

At the time the original protocol was finalized, AZD1775 combined with standard doses of chemotherapeutic agents has been sufficiently well tolerated to permit further evaluations, including evaluations of longer dosing regimens.

Clinical Pharmacokinetics

[redacted]

Potential Drug Interactions

[redacted]

[redacted]

[redacted]

2.4 Correlative Studies Background

During the escalation phase, tumor biopsies will be optional and will be evaluated in pharmacodynamic (PD) studies for evidence of Wee1 inhibition, DNA damage and repair, and apoptosis (γH2AX, pNbs1, Rad51, pTyr15-Cdk, and caspase 3). During the expansion phase, once MTD is reached, mandatory paired tumor biopsies will be pursued in the 6 additional patients enrolled at the MTD to further evaluate PD endpoints and to assess epithelial-mesenchymal transition and immunopharmacodynamics. In Arm B, with a once-daily dosing schedule, tumor biopsies will also be optional during the escalation phase and mandatory during the expansion phase. Paired tumor biopsies will be pursued in up to 20 additional patients enrolled at the maximum tolerated daily dose to further evaluate PD endpoints. Data generated from the tumor biopsies will be essential in establishing drug effect and will be used to inform future sequencing of combination therapies.

Fresh tissue biopsies will be evaluated for pTyr15-Cdk and γH2AX. Further evaluation of markers of DNA damage response will be performed based on tissue availability:

2.4.1 Assessing DNA damage response

Dr. R. Kinders' laboratory, DCTD, has developed a DNA damage response immunofluorescence multiplex assay which includes Rad51, pNbs1, and γH2AX measurements as markers of DNA damage-repair. We additionally plan to measure cleaved caspase 3 as a marker of apoptosis. pTyr15-Cdk expression by immunofluorescence will be evaluated for evidence of target effect.

Histone H2AX is one of the H2A histones present in nucleosomes from normal tissues as well as cancer tissues. H2AX is phosphorylated at its C-terminus (serine 139) within minutes following DNA double-strand breaks. Phosphorylated H2AX, referred to as γH2AX, can be selectively detected using antibodies by Western blots or immunofluorescence. The levels of γH2AX are directly correlated to the amounts of double-strand breaks per cell, and can be used as a dosimeter and biomarker for DNA double-strand breaks.

Nbs1 is an adapter protein, linking Mre11 and Rad50 to form the MRN complex involved in recognition of DNA damage and initiation of the signaling cascade in response to DNA double-strand breaks.

Rad51 is a DNA repair protein involved in homologous recombination, where it facilitates recognition of homologous DNA sequences and catalyzes strand exchange.

Although the effect of AZD1775 as a single agent on the observable amount of DNA repair is not expected to be great, the collected data will be extremely important in evaluating future trials of AZD1775 plus DNA damaging agents. Additionally, γH2AX at longer periods post-dose (>12 hrs) is also a reporter of apoptosis.

Caspase-3 is a key protease activated during early stages of apoptosis via cleavage of its pro-enzyme form into a heterodimer of 17- and 12-kDa subunits. Immunofluorescence assays have been developed and validated by Dr. Kinders' laboratory, and will be performed on biopsy samples as a measure of apoptosis.

CTCs will be collected at baseline and then throughout the study to evaluate whether we can measure changes in the number and phenotype of CTCs in patients over time to explore any correlation with response to treatment or disease progression. This analysis will be performed in Dr. Kinders' lab with the ApoStream instrument, which uses antibody-independent CTC isolation technology that can isolate viable CTCs from epithelial and non-epithelial cancers. The enriched cells are then stained with a panel of antibodies targeting MUC1 (tumor marker) and CEA (tumor marker), CK (epithelial marker), EpCAM (epithelial marker) and β-cat (CTNNB1; epithelial-mesenchymal transition marker) and CD45 (PTPRC; leukocyte marker), as an exclusion marker.

2.4.2 Evaluation of WEE-1 Inhibition with Cancer Immunotherapy

A recent in vitro study demonstrated that WEE-1 inhibition enhances immune mediated lysis of human cancer cells[19]. This was identified when examining the molecular mechanism by which brachyury, a T-box transcription factor that drives the epithelial-mesenchymal transition (EMT) in human carcinoma, imparts resistance to tumor cells. Tumor cells with high levels of brachyury displayed reduced susceptibility to lysis by immune cells, due to inefficient caspase-dependent apoptosis, manifested as inefficient nuclear lamin degradation in the presence of activated effector caspases. This phenomenon correlated with a loss of cell cycle kinase CDK1, which mediates lamin phosphorylation, and pre-treatment of tumor cells with a negative regulator kinase of CDK1, using the WEE-1 inhibitor AZD1775 (MK-1775), countered the defective apoptosis and sensitized resistant tumor cells to immune mediated lysis.

Based on the finding that WEE-1 inhibition enhances immune mediated lysis, we are collaborating with CCR's Laboratory of Tumor Immunology to evaluate the combination of WEE-1 inhibition with cancer immunotherapy, specifically, the effect of AZD1775 on immune peripheral blood mononuclear cells (PBMCs). Peripheral immune cells will be evaluated pre- and post-treatment (baseline and day 8) in both Arm A and Arm B using 27 markers in 4 panels to assess 123 immune subsets (Table 1). These panels identify 9 standard immune cell subsets and 114 markers related to the maturation/function of these cells. The results of this study will determine whether WEE-1 inhibition is immune inert, immune suppressive, or immune stimulatory, and will provide critical information regarding the potential timing and scheduling of combining the AZD1775 with cancer immunotherapy (e.g., therapeutic vaccines or checkpoint inhibitors). Serum will also be collected for analysis of cytokines and chemokines as well as immune activation markers such as CD40L and CD27, and tumor-associated antigens.

Table 1: Planned flow-cytometry analysis of PBMC immune subsets

1.	CD4: Helper T lymphocytes (33 subsets)
2.	CD8: Cytotoxic T lymphocytes (30 subsets)
3.	T-regs: Regulatory T-lymphocytes ($CD4^+$ $CD25^+$ $FoxP3^+$ $CD127^-$) (7 subsets)
4.	B lymphocytes: $CD19^+$ (3 subsets)
5.	NK: Natural killer cells ($CD56^+$ $CD3^-$) (20 subsets)
6.	NK-T: $CD56^+$ $CD3^+$ (4 subsets)
7.	cDCs (Conventional DCs): CD3-CD56-$HLADR^+$CD1c+CD303- (5 subsets)
8.	pDCs (plasmacytoid DCs): $CD3^-CD56^-HLADR^+CD1c^-CD303^+$ (5 subsets)
9.	MDSCs: Myeloid-derived suppressor cells ($CD11b^+$ $HLA\text{-}DR^{low/-}$ $CD33^+$) (16 subsets)

2.4.3 Tumor Immunoscore

The host immune system has been shown to play a role in impeding cancer progression[20]. Therefore, as a pilot correlative study, we will correlate the response to treatment of each patient with colorectal cancer and adenocarcinoma with immunological biomarker expression in tumor biopsy samples by determining the Immunoscore[21]. It is hoped that this information will supplement traditional tumor staging (AJCC-UICC-TNM) which serves to predict patient prognosis by evaluating histopathological characteristics of tumor biopsy samples[22].

Immune cells can infiltrate tumors; their number, type, and location (i.e., immune context) has been associated with disease-free survival and overall survival in large human cancer studies[23-25]. An Immunoscore is determined from the enumeration of a set of these immune cells, for example, cytotoxic ($CD3^+$ and $CD8^+$) and memory ($CD45RO^+$) T-cells in the core of the tumor (CT) and the invasive margin (IM).

Sections from FFPE-tumor tissue blocks will be evaluated for high immune cell infiltration and whether or not they include the CT and IM. Enumeration of lymphocyte populations will be performed by Definiens by immunohistochemistry; clinical validation of the methods is currently being completed.

2.4.4 Epithelial-mesenchymal transition (EMT)

EMT is a dynamic process that leads to increases in the migratory and invasive properties of tumor cells (1). Histologically, these changes involve the loss of epithelial marker E-cadherin (E) and gain of the mesenchymal marker intermediate filament vimentin (V); loss of E-cadherin is linked to poor prognosis, tumor progression, and metastasis[26,27].

Recent data from DCTD confirm the existence of "transitional" cells that co-express both E-cadherin and vimentin at the cellular level in FFPE clinical biopsies. Quantitative analysis of various histologies using an immunofluorescence assay developed and validated by DCTD revealed all possible phenotypes from very epithelial (E+V- colorectal carcinoma), mesenchymal (E-V+ sarcomas), heterogeneous mixtures of E+V- and E-V+ subpopulations, and EMT.

The potential clinical utility of this assay has been demonstrated: treatment with the multikinase inhibitor pazopanib resulted in a significant shift towards the mesenchymal phenotype in drug-sensitive but not insensitive human xenograft models (manuscript in preparation). The EMT immunoassay will be used as to elucidate "baseline" EMT status in study patients—differences observed to date suggest inter-patient phenotypic variation to be as significant a factor as histological differences within carcinomas—as well as to characterize drug response.

2.4.5 Potential predictive molecular biomarkers of AZD1775 antitumor activity

Tumors containing molecular alterations that enhance replicative stress may be particularly susceptible to Wee1 inhibition by AZD1775, with the heightened cell cycle stress in the presence of both factors representing a synthetic lethal interaction. Several genomic alterations that induce replicative stress have been shown to potentially enhance AZD1775 antitumor activity in preclinical and/or clinical studies; these alterations include loss of function mutations in *BRCA1/2*[16,28,29] and *SETD2*[30] and amplification of *CCNE1* or *MYC*[29]. For active Expansion Arm B patients enrolled at the time of Amendment P (11/2/17) approval or thereafter, tumor biopsy cores remaining after completion of PD biomarker analyses will be used for whole-exome and RNA sequencing to identify these and other mutations or gene expression patterns in patient tumor tissue and to examine potential associations between such molecular alterations and AZD1775 antitumor activity.

2.5 Genetic Sequencing Research Ethics

This trial will collect genetic data from active Expansion Arm B patients, enrolled at the time of Amendment P (11/2/17) approval or thereafter, for exploratory studies to examine potential associations between genomic and RNA expression alterations and AZD1775 antitumor activity. Tissue specimens used for sequencing analyses will be de-identified, with only limited clinical annotation (e.g., response to AZD1775) maintained for each specimen. Designing the study poses challenging questions about informed consent, the privacy of the patient and the patient's family, the researchers' obligation to disclose genetic information to the patient, and the use and storage of research data[31-33]. In the vast majority of cases, we do not know the medical significance of genetic variants[34,35]. These challenges will continue to be evaluated to maintain the rigor and integrity of the study and the wellbeing of our patients. A Certificate of Confidentiality has been obtained from the NIH to help protect the privacy of all study participants.

The protocol includes a separate informed consent form for active Expansion Arm B patients enrolled at the time of Amendment P (11/2/17) approval or thereafter; this consent form contains language pertaining to genetic sequencing studies. Active Expansion Arm B patients already enrolled at the time of Amendment P approval will be re-consented to the

new, "genetic analysis" consent form prior to use of their specimens for exploratory sequencing analyses.

Whole-exome sequencing (performed for research purposes but non-CLIA) of tumor and blood can detect non-ambiguous germline variants, which may raise health and privacy implications for the patient and his or her family. WES will not be validated for clinical use, and no clinical decisions can be made based on its results. Furthermore, no genetic data will be shared with the patient.

This study does not meet the criteria specified by the NIH Genomic Data Sharing (GDS) Policy (see Section 12.4), and therefore, a GDS plan has not been included in this protocol.

3 PATIENT SELECTION

3.1 Eligibility Criteria

3.1.1 Patients must have histologically confirmed solid tumors for which all standard therapy known to prolong survival have failed or for which standard therapies do not exist.

3.1.2 Patients must have measurable disease or evaluable disease for the escalation phase; for the 6 additional patients enrolled at MTD for further evaluation of PK and PD endpoints (Expansion Arm A). For the 6-patient BRCA-mutation expansion arm, patients must have measurable disease; however, tumor biopsies are optional. For Expansion Arm B, patients must have tumor amenable to biopsy (excisional or incision biopsies of skin or H & N lesions under visualization) and willingness to undergo a tumor biopsy *or* patient will be undergoing a procedure due to medical necessity during which the tissue may be collected, *or* tumor biopsy tissue from a previous research study or medical care is available for submission at registration. Criteria for the submission of tissue are:

- Tissue must have been collected within 3 months prior to registration
- Patient has not received any intervening therapy for their cancer since the collection of the tumor sample
- Tumor tissue must meet the minimum requirements outlined in Section 9.

3.1.3 Patients must have completed any chemotherapy, radiation therapy, surgery, or biologic therapy ≥ 3 weeks (or ≥ 5 half-lives, whichever is shorter) prior to entering the study. Patients must be ≥ 2 weeks since any prior administration of a study drug in an exploratory IND/Phase 0 study or ≥ 1 week from palliative radiation therapy. Patients must have recovered to eligibility levels from prior toxicity or adverse events.

3.1.4 Age ≥18 years of age.

3.1.5 ECOG performance status ≤1 (Karnofsky >60%, see Appendix A).

3.1.6 Life expectancy of greater than 3 months.

3.1.7 Patients must have normal organ and marrow function as defined below:

– leukocytes	≥3,000/mcL
– absolute neutrophil count	≥1,500/mcL
– platelets	≥100,000/mcL
– hemoglobin	>9 g/dL
– total bilirubin	≤1.5 X institutional upper limit of normal
– AST(SGOT)/	
– ALT(SGPT)	≤3 X institutional upper limit of normal
– creatinine	≤1.5X institutional upper limit of normal
	OR
– creatinine clearance	≥60 mL/min/1.73 m^2 for patients with creatinine levels above institutional normal.

3.1.8 The effects of AZD1775 on the developing human fetus are unknown. For this reason and because molecular inhibitors of Wee1 kinase are known to be teratogenic, women of child-bearing potential (WoCBP) may be included only if acceptable contraception is in place for two weeks before study entry, for the duration of the treatment with the study drug, and for 2 months after the last dose of AZD1775. Male patients who are involved in the study must agree to avoid procreative and unprotected sex (i.e., by using acceptable forms of contraception) and must not donate sperm during the study and for 3 months after the last dose of AZD1775. Where the female partner is pregnant or not using effective birth control, men should be advised to abstain while in the study and for 3 months after the last dose of AZD1775. Female partners, who are of child-bearing potential, of men participating in clinical studies of AZD1775 will also be required to use effective contraceptive measures while their partner is on study drug and for 3 months thereafter. Male patients will be advised to arrange for the freezing of sperm samples prior to the start of the study should they wish to father children while on AZD1775 or during the 3 months after stopping AZD1775.

3.1.9 Because there is an unknown but potential risk for adverse events in nursing infants secondary to treatment of the mother with the study drugs, breastfeeding should be discontinued prior to the first of study drug and women should refrain from nursing throughout the treatment period and for 14 days following the last dose of study drug.

3.1.10 Patients must be able to swallow whole tablets or capsules. Nasogastric or G-tube administration is not allowed. Any gastrointestinal disease which would impair ability to swallow, retain, or absorb drug is not allowed.

3.1.11 Ability to understand and the willingness to sign a written informed consent document.

3.1.12 Patients with prostate cancer can continue to receive treatment with GnRH agonists while on study, as long as there is evidence of disease progression on therapy.

3.2 Exclusion Criteria

3.2.1 Patients who are receiving any other investigational agents.

3.2.2 Patients with known active brain metastases or carcinomatous meningitis are excluded from this clinical trial. Patients whose brain metastatic disease status has remained stable for $\geq$ 4 weeks following treatment of brain metastases are eligible to participate at the discretion of the principal investigator.

3.2.3 Eligibility of subjects receiving any medications or substances with the potential to affect the activity or pharmacokinetics of AZD1775 will be determined following review by the principal investigator.

3.2.4 

3.2.5 Uncontrolled intercurrent illness including, but not limited to, ongoing or active infection, symptomatic congestive heart failure, unstable angina pectoris, cardiac arrhythmia, or psychiatric illness/social situations that would limit compliance with study requirements.

3.2.6 Pregnant women are excluded from this study because the effects of the study drugs on the developing fetus are unknown.

3.2.7 HIV positive patients on antiretroviral therapy are ineligible because of the potential for PK interactions.

3.3 Inclusion of Women and Minorities

Both men and women of all races and ethnic groups are eligible for this trial.

3.4 Eligibility Screening

3.4.1 Histologic confirmation of primary tumor tissue or of known recurrence will be

required from each participant to confirm diagnosis. For the BRCA-mutation expansion arm, confirmation of BRCA-1 or -2 mutations will be required prior to enrollment; pathology reports from outside institutions will be accepted.

3.4.2 History and physical examination: Complete history and physical examination (including height, weight, vital signs, and performance score) will be conducted within 8 days prior to enrollment.

3.4.3 Imaging Studies (Eligibility screening): Every participant should have an evaluation of known sites of disease as part of the eligibility screening. All patients will be required to undergo a CT scan of the chest/abdomen/pelvis to evaluate sites of disease within 28 days prior to enrollment. MRI evaluation of site of disease may be performed in lieu of CT evaluation at the discretion of the principal investigator if it is the opinion of the investigator that this modality would provide a more accurate assessment of disease than a CT would, for a given site.

3.4.4 Laboratory Evaluation: Eligibility screening laboratory data are to be obtained within 8 days prior to enrollment:

- Hematological Profile: CBC with differential.
- Biochemical Profile: albumin, total bilirubin, BUN, calcium, creatinine, phosphorus, SGOT[AST], SGPT[ALT], alkaline phosphatase, chloride, magnesium, potassium, and sodium.
- Coagulation Profile: PT, PTT, INR as clinically indicated prior to tumor biopsy.
- Serum or urine pregnancy test for female participants of childbearing potential.

3.4.5 Cardiac Evaluation: Baseline electrocardiogram will be obtained within 8 days prior to enrollment. Additionally, patients must have electrocardiogram evaluation as clinically indicated.

4 REGISTRATION PROCEDURES

Authorized staff must register an eligible candidate with NCI Central Registration Office (CRO) within 24 hours of signing consent. A registration Eligibility Checklist from the Web site (http://home.ccr.cancer.gov/intra/eligibility/welcome.htm) must be completed and sent via encrypted email to: NCI Central Registration Office (HOIS) (ncicentralregistration-l@mail.nih.gov).

After confirmation of eligibility at Central Registration Office, CRO staff will call pharmacy to advise them of the acceptance of the patient on the protocol prior to the release of any investigational agents. Verification of Registration will be forwarded electronically via e-mail. Please note that it is very important for all registrars to acquire encrypted e-mail from NIH Help Desk, since the verification of registration includes patient's information. A recorder is available during non-working hours.

Off Protocol Therapy and Off-Study Procedure: Authorized staff must notify the Central Registration Office (CRO) when a patient is taken off protocol therapy and when a patient is taken off-study. A Participant Status Updates Form from the website (http://home.ccr.cancer.gov/intra/eligibility/welcome.htm) main page must be completed and sent via encrypted email to: NCI Central Registration Office (HOIS) (ncicentralregistration-l@mail.nih.gov).

5 TREATMENT PLAN

This is an open-label Phase I trial evaluating safety and tolerability of single-agent AZD1775 (MK-1775), an oral inhibitor of Wee1, in patients with refractory advanced solid tumors. Reported adverse events and potential risks for AZD1775 are described in Section 7. Appropriate dose modifications for AZD1775 are described in Section 6.

Patient evaluations will be performed throughout the study as described below. Baseline history, physical examination, laboratory evaluations, and ECG must be conducted within 8 days prior to start of protocol therapy. If protocol therapy is started within 8 days of the eligibility screening evaluations (see Section 3.4), the results from these screening evaluations may be used as baseline measurements. If >8 days have passed since the screening evaluations, the medical history, physical examination, laboratory evaluations, and ECG must be repeated prior to starting protocol therapy. Baseline tumor imaging must be performed within 28 days prior to start of protocol therapy. If protocol therapy is started within 28 days of the eligibility screening tumor imaging, the screening evaluation imaging results may be used as baseline measurements; if >28 days have passed since the screening evaluation tumor imaging, the imaging must be repeated prior to starting protocol therapy.

For dose level 1 of the original dosing schema (Arm A), AZD1775 will be administered orally for 5 doses each cycle, with doses administered approximately 12 hours apart starting with the first dose for that week (for example, if the patient takes the first dose at 9 am on Monday then the next dose will be administered at 9 pm that evening). Starting at dose level 2 and onwards in this arm, AZD1775 will be administered orally for 5 doses for the first 2 weeks of each cycle, with doses administered approximately 12 hours apart starting with the first dose for that week (as explained above except it will occur over the first two weeks of the cycle). Each cycle is 21 days (± 1 day for scheduling). Patients will be asked to maintain a Study Medication Diary (Appendix D) and record each dose of medication. Patients will be given instructions for completing the medication diary and will be asked to return it to the clinic staff at the end of each cycle.

Once the MTD is established, 6 additional patients will be enrolled at the MTD to further evaluate that dose for PK and PD endpoints (Expansion Arm A). An additional expansion arm of 6 patients with documented tumors harboring BRCA-1 or -2 mutations will also be enrolled at the MTD to further explore the safety of AZD1775 and obtain preliminary evidence of activity in this patient population. This information may inform patient selection for further development of this agent. Patients in this BRCA-mutant expansion arm must have measurable disease; tumor biopsies will be optional.

Based on preclinical data with once daily AZD1775 dosing, Arm B, starting at 200 mg once daily for 5 doses each week, for two consecutive weeks will be established to evaluate PD and PK endpoints. Patients will be accrued in arms of 3, based on a 3+3 design, for up to two dose levels. The cycle length will remain at 21 days (+ 1 day for scheduling). Once the maximum tolerated single daily dosing is established, up to 20 additional patients will be enrolled on Expansion Arm B with mandatory biopsies to further evaluate PD endpoints.

History and physical examination will be done at baseline (within 8 days prior to the start of protocol therapy) and before the start of each new cycle thereafter (up to 3 days before the start of a new cycle). Patients will be examined on D1 and D8 at the clinical center during the first cycle and prior to every new cycle.

As of Amendment G (dated 07/17/2014), the MTD for Arm A was established at 225 mg twice daily for 5 doses each week, for two consecutive weeks. As of Amendment M (dated 11/20/16), the Arm B MTD was established at 300 mg once daily on days 1-5 each week, for two consecutive weeks.

Labs (CBC with differential; serum chemistries) will be performed at baseline (within 8 days prior to the start of protocol therapy), D8, and D15 of cycle 1 (±1 day due to scheduling conflicts), then at the start of each cycle (up to 3 days before start of a new cycle).

CT scans will be performed at baseline (within 8 days prior to the start of protocol therapy), and repeat imaging scans will be performed every 2 cycles (3 or 4 cycles for patients on study for more than one or three years, respectively). MRI evaluation of site of disease may be performed in lieu of CT evaluation at the discretion of the principal investigator if it is the opinion of the investigator that this modality would provide a more accurate assessment of disease than a CT would for a given site.

5.1 Agent Administration

After cycle 2, a cycle will be considered completed if 90% of the prescribed AZD1775 doses are administered.

5.1.1 Dose Escalation Phase (Arm A)

The primary objective of the trial is to establish the safety, tolerability, and maximum tolerated dose of single-agent AZD1775.

For dose level 1, AZD1775 will be administered orally for 5 doses each cycle, with doses administered approximately 12 hours apart starting with the first dose for that week (for example, if the patient takes the first dose at 9AM on Monday then the next dose will be administered at 9 pm that evening). Starting at dose level 2 and onwards, AZD1775 will be administered orally for 5 doses for the first 2 weeks of each cycle, with doses administered approximately 12 hours apart starting with the first dose for that week (as explained above except it will occur over the first two weeks of the cycle). The duration of a cycle will be 21 days

(± 1 day for scheduling). Dose escalation will proceed in arms of 3 patients each (3+3 design) as outlined below (Table 1):

Table 1. Arm A Dose Escalation Scheme

Dose Escalation Schedule for Arm A	
Dose Level	***AZD1775 (orally BID) x 5 doses/weeks***
-1	200 mg BID x 5 doses (days 1-3) for 1 week
1	225 mg BID x 5 doses (days 1-3) for 1 week
2	225 mg BID x 5 doses (days 1-3 each week) for 2 weeks
3	300 mg BID x 5 doses (days 1-3 each week) for 2 weeks
4	400 mg BID x 5 doses (days 1-3 each week) for 2 weeks
5	550 mg BID x 5 doses (days 1-3 each week) for 2 weeks

Optional paired tumor biopsies will be obtained in patients who have disease amenable to biopsy during the escalation phase; mandatory paired tumor biopsies will be pursued in patients during the expansion phase in the 6 additional patients enrolled at the MTD (Expansion Arm A). For the 6-patient BRCA-mutant expansion arm at the MTD, disease must be measurable; however, tumor biopsies will be optional.

The trial will follow a standard 3+3 dose escalation study wherein 3 patients should have completed at least one cycle of therapy prior to considering dose escalation in the next arm of patients. Dose escalation will proceed until dose-limiting toxicity is observed. DLT is defined in Section 5.2. Determination of DLT will be based on toxicities observed in the first cycle of therapy. Patients are considered evaluable for toxicity for the purpose of arm dose escalation decisions if they either 1) experienced DLT or 2) have received 90% of treatment doses for cycle one of therapy and have been followed for one full cycle without DLT. All toxicities will be reported for all patients who receive any amount of study drug on this study. Evaluation of toxicity will begin with study drug administration on cycle 1 day 1.

Intra-patient dose escalation will be permitted if: **a)** there is no toxicity > Grade 1 that is related (possibly, probably, or definitely) to the study drug at the initial dose level experienced by that patient, **b)** higher doses have been evaluated and completed without DLT, and **c)** the patient's disease has not progressed. Doses may be escalated, provided conditions a–c are met, up to the last evaluated dose level NOT associated with DLT.

5.1.2 Dose Escalation Phase (Arm B)

For Arm B, AZD1775 will be administered once daily for 5 doses each week, for two consecutive weeks. The duration of a cycle will be 21 days (+1 day for scheduling). Dose escalation will proceed in arms of 3 patients each (3+3 design) as outlined in Table 2:

Table 2. Arm B Escalation Scheme

Dose Escalation Schedule for Arm B	
Dose Level	AZD1775 (orally once daily)
-1	200 mg daily D1-3 each week for 2 weeks
1	200 mg daily x D1-5 each week for 2 weeks
2	225 mg daily x D1-5 each week for 2 weeks
3	250 mg daily D1-5 each week for 2 weeks
4	300 mg daily D1-5 each week for 2 weeks
5	400 mg daily D1-5 each week for 2 weeks
6	500 mg daily D1-5 each week for 2 weeks

5.2 Definition of Dose-Limiting Toxicity

Determination of dose-limiting toxicity will be based on toxicities observed in the first cycle of therapy. Dose escalation will proceed within each arm according to the following scheme. Dose-limiting toxicity (DLT) is defined below.

Number of Patients with DLT at a Given Dose Level	Escalation Decision Rule
0 out of 3	Enter 3 patients at the next dose level.
≥2	Dose escalation will be stopped. This dose level will be declared the maximally administered dose (highest dose administered). Three (3) additional patients will be entered at the next lowest dose level if only 3 patients were treated previously at that dose.
1 out of 3	Enter at least 3 more patients at this dose level. • If 0 of these 3 patients experience DLT, proceed to the next dose level. • If 1 or more of this group suffer DLT, then dose escalation is stopped, and this dose is declared the maximally administered dose. Three (3) additional patients will be entered at the next lowest dose level if only 3 patients were treated previously at that dose.
≤1 out of 6 at highest dose level below the maximally administered dose	This is generally the recommended phase 2 dose. At least 6 patients must be entered at the recommended phase 2 dose.

Dose limiting toxicity (DLT) is defined as an adverse event that is related (possibly, probably, or definitely) to administration of AZD1775 and fulfills one of the following criteria:

5.2.1 Grade ≥ 3 Non-Hematological Toxicity

Grade ≥ 3 non-hematological toxicity felt to be related to study medications will be considered dose-limiting with the following clarifications:

5.2.1.1 Diarrhea Grade 3 will only be considered dose-limiting if it is refractory to treatment as outlined in Section 5.3.2, Supportive Care Guidelines, and unable to be corrected to Grade 2 or less within 24 hours. Bloody or Grade 4 diarrhea will be dose-limiting.
5.2.1.2 Nausea and vomiting Grade 3 will only be considered dose-limiting if it is refractory to anti-emetic therapy and unable to be corrected to Grade 1 or less within 24 hours (Section 5.3.1).
5.2.1.3 Rise in creatinine to Grade 3, not corrected to Grade 1 or less within 48 hours with IV fluids will be considered dose-limiting. All Grade 4 rises in creatinine will be dose limiting.
5.2.1.4 Grade $\geq$ 3 metabolic toxicities unable to be corrected to Grade 1 or baseline within 48 hours (hypocalcemia or hypercalcemia, hypomagnesemia or hypermagnesemia, and hyponatremia) will be considered dose limiting. For hypokalemia or hyperkalemia, grade $\geq$ 2 toxicities unable to be corrected to grade 1 or less within 48 hours will be considered dose limiting. Grade 4 metabolic toxicities that are symptomatic will be considered dose-limiting regardless of duration or ability to correct.

5.2.2 Grade 4 thrombocytopenia or Grade 3 thrombocytopenia associated with bleeding.

5.2.3 Grade 4 neutropenia $\geq$ 5 days or febrile neutropenia.

5.2.4 Any degree of anemia, leukopenia in the absence of grade 4 neutropenia $\geq$ 4 days, or lymphopenia will not be considered dose limiting.

5.2.5 Any degree of alopecia will not be considered dose limiting.

5.2.6 Grade 3 fatigue of greater than 1 week duration.

5.2.7 Failure to tolerate 100% of the dosing in the first cycle will be considered a DLT.

5.3 General Concomitant Medication and Supportive Care Guidelines

All patients will be provided with the best available supportive care. All concurrent medications should be documented prior to initiation of treatment, and be periodically reviewed with the patient.

[redacted]

5.3.1 Nausea/Vomiting

Anti-emetics will not be administered routinely prior to AZD1775. However, if a patient develops nausea/vomiting, anti-emetics such as but not limited to prochlorperazine, metoclopramide, 5-HT3 antagonists may be given. Aprepitant

and fosaprepitant are not permitted due to known drug interactions with AZD1775. In addition, if a patient develops nausea and/or vomiting that is Grade 2 or greater, antiemetics may be instituted prophylactically at the discretion of the investigator. Nausea and vomiting will be considered refractory if it does not resolve to≤ Grade 1 with treatment with a combination of at least 2 of the antiemetics within 24 hours. Patients should be strongly encouraged to maintain liberal oral fluid intake.

5.3.2 Diarrhea

If diarrhea develops and does not have an identifiable cause other than study drug administration, anti-diarrheals such as Lomotil (diphenoxylate HCl 2.5 mg + atropine sulfate 0.025 mg/tablet) dosed according to package insert or loperamide 4 mg po after the first unformed stool with 2 mg po with every 2 hours as long as unformed stools continue (4 mg every 4 hours while asleep). No more than 16 mg of loperamide should be taken during a 24-hour period. This regimen can be repeated for each diarrheal episode. Diarrhea will be considered refractory if it does not resolve within 24 hours ≤ to Grade 2 with the above regimen (maximum of 16 mg of loperamide in a 24-hour period). If the patient develops blood or mucus in the stool, dehydration, or hemodynamic instability, or fever along with the diarrhea, anti-diarrheals will be discontinued and the patient will be treated with IV fluids and antibiotics as medically indicated.

5.3.3 Neutropenia

To reduce the risk of severe myelosuppression events, a complete blood count (CBC) should be performed weekly during cycle 1, and at the start of each subsequent cycle (up to 3 days before start of new cycle) in the dose escalation phase. In the expansion phase, CBC will be performed on D1 and D8 of cycle 1, then at the start of each cycle (up to 3 days before start of new cycle). Febrile neutropenia is a life-threatening complication requiring hospitalization and urgent broad spectrum antibiotics, as well as an aggressive search for the source and microbial cause of the episode. Growth factors to prevent neutropenia will not be administered prophylactically. If necessary, they may be administered according to accepted American Society of Clinical Oncology (ASCO) guidelines to allow re-treatment.

5.3.4 Anemia

Symptomatic anemia should be treated with red blood cell transfusion and is recommended if the hemoglobin falls below 8 g/dL. The initiation of erythropoietic therapy for the management of chemotherapy-induced anemia follows the American Society of Hematology/ASCO clinical practice guidelines (http://www.asco.org).

5.3.5 Thrombocytopenia

Thrombocytopenia will be treated conservatively. In the absence of bleeding, or a necessary invasive procedure, platelet transfusions should be given for a platelet

count ≤10,000/mm^3. If invasive procedure(s) is (are) planned, or the patient develops bleeding, platelet transfusions should be administered in accordance with the standard of practice, usually maintaining a platelet count above 50,000/mm^3.

5.4 Duration of Therapy

In the absence of treatment delays due to adverse events, treatment may continue until one of the following criteria applies:

- Disease progression
- Intercurrent illness that prevents further administration of treatment
- Significant toxicity occurs despite 2 dose reductions as described in Section 6 or no lower dose level exists
- Pregnancy
- Patient decides to withdraw from the study, or
- General or specific changes in the patient's condition render the patient unacceptable for further treatment in the judgment of the investigator

5.5 Duration of Follow Up

Patients will be followed for 30 days after the last dose is administered or until one of the following occurs: patient enrolls on another protocol, patient receives standard of care, or death, whichever comes first. The follow-up will consist of a phone call between Days 27-30 after the last dose to evaluate adverse events that were ongoing and any new events that might be deemed related to the therapy. Toxicities felt to be possibly, probably, or definitely related to the study drugs that have not resolved or stabilized by Day 30 post-treatment will be followed until stabilization or resolution via phone calls as clinically indicated. An optional biopsy from patients who have progressed may be requested if the patient has not yet received any new treatment to assess whether progression correlates with a change in study pharmacodynamic marker; separate consent will be obtained.

5.6 Criteria for Removal from Study

Patients will be removed from study for one of the following reasons: completed 30-day follow up period or toxicities are unresolved but stabilized, patient enrolls on another protocol, or patient receives standard of care. The reason for study removal and the date the patient was removed must be documented in the medical record and communicated to Central Registration per Section 4.

6 DOSING DELAYS/DOSE MODIFICATIONS

Toxicities should have resolved to ≤ Grade 2 prior to starting the next cycle. Treatment may be delayed for a maximum of 2 weeks beyond the actual cycle length of 21 days for toxicities that develop and do not resolve as defined above. Beyond two weeks, the patient will not receive further therapy on this protocol and will be followed for resolution of toxicities. Treatments may be delayed up to 7 days past the end of the previous cycle of 21 days for scheduling conflicts at the discretion of the investigator.

Patients will be allowed up to 2 dose reductions. If more than 2 dose reductions are required, the patient will be removed from the study.

6.1 Dose Reduction

Dose modifications are defined below:

6.1.1 Grade 2 Drug-related toxicity: No changes will be made to the dose of AZD1775 for Grade 2 toxicities. Therapy will not be interrupted for Grade 2 hematologic toxicities.

6.1.2 Grade 3-4 Drug-related non-hematologic toxicities: Doses of AZD1775 will be held until toxicities recover to ≤ Grade 2 or baseline prior to re-initiating treatment at the next lower dose level. Electrolyte abnormalities will not require dose reduction if resolution to Grade 2 or less is documented within 72 hours. Dose modifications for nausea, vomiting, and diarrhea will be made only if they are refractory to treatment (See Section 5.3).

6.1.3 Grade 3 Drug-related hematologic toxicities: Dose of AZD1775 will be held until hematologic toxicities, except anemia, lymphopenia, or leukopenia in the absence of neutropenia, have resolved to ≤ Grade 2 prior to re-initiating treatment at the same dose level.

6.1.4 Grade 4 Drug-related Hematologic Toxicities: Dose of AZD1775 will be held until hematologic toxicities, except anemia, lymphopenia, or leukopenia in the absence of neutropenia, have resolved to ≤ Grade 2 prior to re-initiating treatment at the next lower dose level.

6.2 Definition of Complete Course

A cycle is defined as 21 days, with D1 starting the first day of AZD1775 administration. If a patient receives 90% of scheduled treatments of AZD1775 and remains in the study until D21, the patient will be considered to have completed a cycle of therapy. Patients who do not complete a cycle of therapy for reasons other than toxicity will be replaced. Patients who do not complete one cycle of therapy due to toxicity will not be replaced. All patients should have completed one cycle of therapy (unless removed from the study due to toxicity), at which point they will be evaluable.

7 ADVERSE EVENTS: LIST AND REPORTING REQUIREMENTS

Adverse event (AE) monitoring and reporting is a routine part of every clinical trial. The following list of AEs (Section 7.1) and the characteristics of an observed AE (Section 7.2) will determine whether the event requires expedited reporting (via CTEP-AERS) **in addition** to routine reporting.

7.1 Comprehensive Adverse Events and Potential Risks list (CAEPR)

The Comprehensive Adverse Events and Potential Risks list (CAEPR) provides a single list of reported and/or potential adverse events (AE) associated with an agent using a uniform presentation of events by body system. In addition to the comprehensive list, a subset, the Specific Protocol Exceptions to Expedited Reporting (SPEER), appears in a separate column and is identified with bold and italicized text. This subset of AEs (SPEER) is a list of events that are protocol specific exceptions to expedited reporting to NCI (except as noted below). Refer to the 'CTEP, NCI Guidelines: Adverse Event Reporting Requirements' http://ctep.cancer.gov/protocolDevelopment/electronic_applications/docs/aeguidelines.pdf for further clarification. *Frequency is provided based on 313 patients.* Below is the CAEPR for AZD1775 (adavosertib).

NOTE: Report AEs on the SPEER **ONLY IF** they exceed the grade noted in parentheses next to the AE in the SPEER. If this CAEPR is part of a combination protocol using multiple investigational agents and has an AE listed on different SPEERs, use the lower of the grades to determine if expedited reporting is required.

Version 2.6, May 14, 2019[1]

[redacted]

[redacted]

[1]This table will be updated as the toxicity profile of the agent is revised. Updates will be distributed to all Principal Investigators at the time of revision. The current version can be obtained by contacting PIO@CTEP.NCI.NIH.GOV. Your name, the name of the investigator, the protocol and the agent should be included in the e-mail.

[2]Gastrointestinal hemorrhage includes Anal hemorrhage, Cecal hemorrhage, Colonic hemorrhage, Duodenal hemorrhage, Esophageal hemorrhage, Esophageal varices hemorrhage, Gastric hemorrhage, Hemorrhoidal hemorrhage, Ileal hemorrhage, Intra-abdominal hemorrhage, Jejunal hemorrhage, Lower gastrointestinal hemorrhage, Oral hemorrhage, Pancreatic hemorrhage, Rectal hemorrhage, Retroperitoneal hemorrhage, and Upper gastrointestinal hemorrhage under the GASTROINTESTINAL DISORDERS SOC.

[3]Infection includes all 75 sites of infection under the INFECTIONS AND INFESTATIONS SOC.

[4]Rash may include rash, erythema, eczema, and rash maculo-papular.

[5]Peripheral neuropathy includes both peripheral motor neuropathy and peripheral sensory neuropathy.

[6]Acute kidney injury includes renal impairment and acute renal insufficiency.

[redacted]

BLOOD AND LYMPHATIC SYSTEM DISORDERS - Blood and lymphatic system disorders - Other (pancytopenia); Blood and lymphatic system disorders - Other (thrombocytosis); Leukocytosis
CARDIAC DISORDERS - Cardiac disorders - Other (cardiomegaly); Chest pain - cardiac; Myocardial infarction; Palpitations; Sinus bradycardia; Sinus tachycardia
EAR AND LABYRINTH DISORDERS - Ear pain; Hearing impaired; Tinnitus
EYE DISORDERS - Blurred vision; Cataract; Eye disorders - Other (eye swelling); Eye pain; Keratitis; Photophobia; Scleral disorder; Vision decreased; Watering eyes
GASTROINTESTINAL DISORDERS - Abdominal distension; Anal pain; Ascites; Belching; Bloating; Cheilitis; Colitis; Colonic obstruction; Dry mouth; Duodenal ulcer; Dysphagia; Enterocolitis; Flatulence; Gastric ulcer; Gastritis; Hemorrhoids; Oral pain; Rectal pain; Small intestinal obstruction
GENERAL DISORDERS AND ADMINISTRATION SITE CONDITIONS - Chills; Death NOS; Edema trunk; Flu like symptoms; Gait disturbance; General disorders and administration site conditions - Other (catheter site pain); Infusion site extravasation; Malaise; Non-cardiac chest pain; Pain
IMMUNE SYSTEM DISORDERS - Allergic reaction; Anaphylaxis; Cytokine release syndrome
INJURY, POISONING AND PROCEDURAL COMPLICATIONS - Fall; Injury, poisoning and procedural complications - Other (excoriation); Injury, poisoning and procedural complications - Other (ligament sprain)
INVESTIGATIONS - Alkaline phosphatase increased; Aspartate aminotransferase increased; Blood bilirubin increased; Creatinine increased; GGT increased; Investigations - Other (blood urea increased); Lymphocyte count increased; Weight loss
METABOLISM AND NUTRITION DISORDERS - Alkalosis; Hypercalcemia; Hyperglycemia; Hyperkalemia; Hyperuricemia; Hypoalbuminemia; Hypocalcemia; Hyponatremia; Hypophosphatemia; Tumor lysis syndrome
MUSCULOSKELETAL AND CONNECTIVE TISSUE DISORDERS - Arthralgia; Arthritis; Bone pain; Flank pain; Generalized muscle weakness; Muscle cramp; Muscle weakness lower limb; Musculoskeletal and connective tissue disorder - Other (groin pain); Neck pain; Pain in extremity
NEOPLASMS BENIGN, MALIGNANT AND UNSPECIFIED (INCL CYSTS AND POLYPS) - Neoplasms benign, malignant and unspecified (incl cysts and polyps) - Other (carcinoid tumor); Tumor pain

NERVOUS SYSTEM DISORDERS - Central nervous system necrosis; Cognitive disturbance; Dysesthesia; Dysgeusia; Encephalopathy; Lethargy; Nervous system disorders - Other (hemiparesis); Paresthesia; Peripheral neuropathy[5]; Presyncope; Somnolence; Syncope
PSYCHIATRIC DISORDERS - Agitation; Anxiety; Confusion; Depression
RENAL AND URINARY DISORDERS - Acute kidney injury[6]; Hematuria; Urinary frequency; Urinary incontinence; Urinary retention; Urinary tract pain
REPRODUCTIVE SYSTEM AND BREAST DISORDERS - Genital edema; Reproductive system and breast disorders - Other (female genital tract fistula)
RESPIRATORY, THORACIC AND MEDIASTINAL DISORDERS - Allergic rhinitis; Bronchopulmonary hemorrhage; Epistaxis; Hiccups; Nasal congestion; Pleural effusion; Pneumonitis; Pulmonary hypertension; Respiratory, thoracic and mediastinal disorders - Other (diaphragmalgia); Voice alteration; Wheezing
SKIN AND SUBCUTANEOUS TISSUE DISORDERS - Alopecia; Bullous dermatitis; Dry skin; Hyperhidrosis; Pain of skin; Palmar-plantar erythrodysesthesia syndrome; Pruritus; Purpura; Rash acneiform; Skin ulceration; Urticaria
VASCULAR DISORDERS - Flushing; Hematoma; Hot flashes; Hypertension; Hypotension; Thromboembolic event

Note: AZD1775 (adavosertib) in combination with other agents could cause an exacerbation of any adverse event currently known to be caused by the other agent, or the combination may result in events never previously associated with either agent.

7.2 Adverse Event Characteristics

- **CTCAE term (AE description) and grade:** The descriptions and grading scales found in the revised NCI Common Terminology Criteria for Adverse Events (CTCAE) version 4.0 will be utilized for AE reporting until March 31, 2018. CTCAE version 5.0 will be utilized for AE reporting beginning April 1, 2018. All appropriate treatment areas should have access to a copy of the CTCAE version 5.0. A copy of the CTCAE version 5.0 can be downloaded from the CTEP web site http://ctep.cancer.gov/protocolDevelopment/electronic_applications/ctc.htm.

- **'Expectedness'**: AEs can be 'Unexpected' or 'Expected' (see Section 7.1 above) for expedited reporting purposes only. 'Expected' AEs (the ASAEL) are ***bold and italicized*** in the CAEPR (Section 7.1).

- **Attribution** of the AE:
  1. Definite – The AE *is clearly related* to the study treatment.
  2. Probable – The AE *is likely related* to the study treatment.
  3. Possible – The AE *may be related* to the study treatment.
  4. Unlikely – The AE *is doubtfully related* to the study treatment.
  5. Unrelated – The AE *is clearly NOT related* to the study treatment.

7.3 Expedited Adverse Event Reporting

7.3.1 Expedited AE reporting for this study must use CTEP-AERS (CTEP Adverse Event Reporting System), accessed via the CTEP Web site (http://ctep.cancer.gov). The reporting procedures to be followed are presented in the "NCI Guidelines for Investigators: Adverse Event Reporting Requirements for DCTD (CTEP and CIP) and DCP INDs and IDEs" which can be downloaded from the CTEP Web site (http://ctep.cancer.gov). These requirements are briefly outlined in the tables below (Section 7.3.3).

7.3.2 In the rare occurrence when Internet connectivity is lost, a 24-hour notification is to be made to CTEP by telephone at 301-897-7497. Once Internet connectivity is restored, the 24-hour notification phoned in must be entered electronically into CTEP-AERS by the original submitter at the site.

7.3.3 Expedited Reporting Guidelines

Use the NCI protocol number and the protocol-specific patient ID assigned during trial registration on all reports.

Note: A death on study requires both routine and expedited reporting regardless of causality, unless as noted below. Attribution to treatment or other cause must be provided.

Death due to progressive disease should be reported as **Grade 5 "Disease Progression"** in the system organ class (SOC) "General disorders and administration site conditions." Evidence that the death was a manifestation of underlying disease (e.g., radiological changes suggesting tumor growth or progression: clinical deterioration associated with a disease process) should be submitted.

Phase I and Early Phase I Studies: Expedited Reporting Requirements for Adverse Events that Occur on Studies under an IND/IDE within 30 Days of the Last Administration of the Investigational Agent/Intervention [1, 2]

FDA REPORTING REQUIREMENTS FOR SERIOUS ADVERSE EVENTS (21 CFR Part 312)

NOTE: Investigators **MUST** immediately report to the sponsor (NCI) **ANY** Serious Adverse Events, whether or not they are considered related to the investigational agent(s)/intervention (21 CFR 312.64)

An adverse event is considered serious if it results in **ANY** of the following outcomes:

1) Death
2) A life-threatening adverse event
3) An adverse event that results in inpatient hospitalization or prolongation of existing hospitalization for ≥ 24 hours
4) A persistent or significant incapacity or substantial disruption of the ability to conduct normal life functions
5) A congenital anomaly/birth defect.
6) Important Medical Events (IME) that may not result in death, be life threatening, or require hospitalization may be considered serious when, based upon medical judgment, they may

jeopardize the patient or subject and may require medical or surgical intervention to prevent one of the outcomes listed in this definition. (FDA, 21 CFR 312.32; ICH E2A and ICH E6).

ALL SERIOUS adverse events that meet the above criteria MUST be immediately reported to the NCI via CTEP-AERS within the timeframes detailed in the table below.

Hospitalization	Grade 1 and Grade 2 Timeframes	Grade 3-5 Timeframes
Resulting in Hospitalization ≥ 24 hrs	10 Calendar Days	24-Hour 5 Calendar Days
Not resulting in Hospitalization ≥ 24 hrs	Not required	

NOTE: Protocol specific exceptions to expedited reporting of serious adverse events are found in the Specific Protocol Exceptions to Expedited Reporting (SPEER) portion of the CAEPR.

Expedited AE reporting timelines are defined as:

- "24-Hour; 5 Calendar Days" - The AE must initially be reported via CTEP-AERS within 24 hours of learning of the AE, followed by a complete expedited report within 5 calendar days of the initial 24-hour report.
- "10 Calendar Days" - A complete expedited report on the AE must be submitted within 10 calendar days of learning of the AE.

[1]Serious adverse events that occur more than 30 days after the last administration of investigational agent/intervention and have an attribution of possible, probable, or definite require reporting as follows:

Expedited 24-hour notification followed by complete report within 5 calendar days for:

- All Grade 3, 4, and Grade 5 AEs

Expedited 10 calendar day reports for:

- Grade 2 AEs resulting in hospitalization or prolongation of hospitalization

[2] For studies using PET or SPECT IND agents, the AE reporting period is limited to 10 radioactive half-lives, rounded UP to the nearest whole day, after the agent/intervention was last administered. Footnote "1" above applies after this reporting period.

Effective Date: May 5, 2011

7.3.4 Protocol-specific expedited AE reporting exclusions

For this protocol only, certain AEs/grades are exceptions to the Expedited Reporting Guidelines and do not require expedited reporting (i.e., CTEP-AERS). These are: any grade lymphopenia, any grade alopecia, Grade 2 electrolyte (sodium, potassium, phosphorous, magnesium) abnormalities, Grade 2 anemia, Grade 2 hypoalbuminemia, Grade 2 hyperglycemia, Grade 2 INR, Grade 2 PTT, Grade 2 hyperglycemia, and Grade 2 hyperuricemia will NOT be reported through CTEP-AERS but will be reported in the routine data submissions.

7.3.5 Pregnancy, Fetal Death, and Death Neonatal

NOTE: When submitting CTEP-AERS reports for "Pregnancy", "Pregnancy loss", or "Neonatal loss", the Pregnancy Information Form should be completed for patients who became pregnant on study, and faxed along with any additional medical information to **(301) 230-0159.** The potential risk of exposure of the fetus to the investigational agent(s) or chemotherapy agent(s) should be documented in the "Description of Event" section of the CTEP-AERS report.

7.3.5.1 Pregnancy

- Because patients who become pregnant on study risk intrauterine exposure of the fetus to agents which may be teratogenic DCTD/DCP is requesting that pregnancy should be reported in an expedited manner via CTEP-AERS as Grade 3 "***Pregnancy, puerperium and perinatal conditions - Other (pregnancy)***" under the ***Pregnancy, puerperium and perinatal conditions*** SOC.
- The pregnancy outcome for patients on study should be reported via CTEP-AERS at the time the outcome becomes known, accompanied by the same Pregnancy Report Form used for the initial report.

7.3.5.2 Pregnancy loss

- Pregnancy loss is defined in CTCAE as "Death in utero."
- Any pregnancy loss should be reported expeditiously, as Grade 4 "Pregnancy loss" under the Pregnancy, puerperium and perinatal conditions SOC.
- A pregnancy loss should NOT be reported as a Grade 5 event under the Pregnancy, puerperium and perinatal conditions SOC, as currently CTEP-AERS recognizes this event as a patient death.

7.3.5.3 Death Neonatal

- Neonatal death, defined in CTCAE as "A disorder characterized by cessation of life occurring during the first 28 days of life" that is felt by the investigator to be at least possibly due to the investigational agent/intervention, should be reported expeditiously.
- A neonatal death should be reported expeditiously as Grade 4 "Death neonatal" under the General disorders and administration SOC.
- Neonatal death should NOT be reported as Grade 5 "Death neonatal" under the General disorders and administration SOC. If reported as such, the CTEP-AERS interprets this as a death of the patient being treated.

7.3.6 NIH IRB Expedited Reporting of Adverse Events, Unanticipated Problems, and Deaths

7.3.6.1 Definitions

Adverse event

An adverse event is defined as any reaction, side effect, or untoward event that occurs during the course of the clinical trial associated with the use of a drug in humans, whether or not the event is considered related to the treatment or clinically significant. For this study, AEs will include events reported by the patient, as well as clinically significant abnormal findings on physical examination or laboratory evaluation. A new illness, symptom, sign or clinically significant laboratory abnormality or worsening of a pre-existing condition or abnormality is considered an AE. All AEs must be recorded on the AE case report form unless otherwise noted in Section 7.3.4.

All AEs, including clinically significant abnormal findings on laboratory evaluations, regardless of severity, will be followed until satisfactory resolution. AEs should be reported up to 30 days following the last dose of study drug. An abnormal laboratory value will be considered an AE if the laboratory abnormality is characterized by any of the following:

- Results in discontinuation from the study
- Is associated with clinical signs or symptoms
- Requires treatment or any other therapeutic intervention
- Is associated with death or another serious adverse event, including hospitalization
- Is judged by the Investigator to be of significant clinical impact
- If any abnormal laboratory result is considered clinically significant, the investigator will provide details about the action taken with respect to the test drug and about the patient's outcome.

Suspected adverse reaction

Suspected adverse reaction means any adverse event for which there is a *reasonable possibility* that the drug caused the adverse event. For the purposes of IND safety reporting, 'reasonable possibility' means there is evidence to suggest a causal relationship between the drug and the adverse event. A suspected adverse reaction implies a lesser degree of certainty about causality than adverse reaction, which means any adverse event caused by a drug.

Unexpected adverse reaction

An adverse event or suspected adverse reaction is considered "unexpected" if it is not listed in the investigator brochure or is not listed at the specificity or severity that has been observed; or, if an investigator brochure is not required or available, is not consistent with the risk information described in the general investigational plan or elsewhere in the current application. "Unexpected" also refers to adverse events or suspected adverse reactions that are mentioned in the investigator

brochure as occurring with a class of drugs or as anticipated from the pharmacological properties of the drug, but are not specifically mentioned as occurring with the particular drug under investigation.

Serious
An Unanticipated Problem or Protocol Deviation is serious if it meets the definition of a Serious Adverse Event or if it compromises the safety, welfare or rights of subjects or others.

Serious Adverse Event
An adverse event or suspected adverse reaction is considered serious if in the view of the investigator or the sponsor, it results in any of the following:

- Death,
- A life-threatening adverse drug experience
- Inpatient hospitalization or prolongation of existing hospitalization
- Persistent or significant incapacity or substantial disruption of the ability to conduct normal life functions
- A congenital anomaly/birth defect.
- Important medical events that may not result in death, be life-threatening, or require hospitalization may be considered a serious adverse drug experience when, based upon appropriate medical judgment, they may jeopardize the patient or subject and may require medical or surgical intervention to prevent one of the outcomes listed in this definition.

Disability
A substantial disruption of a person's ability to conduct normal life functions.

Life-threatening adverse drug experience
Any adverse event or suspected adverse reaction that places the patient or subject, in the view of the investigator or sponsor, at immediate risk of death from the reaction as it occurred, i.e., it does not include a reaction that had it occurred in a more severe form, might have caused death.

Protocol Deviation (NIH Definition)
Any change, divergence, or departure from the IRB approved research protocol.

Non-compliance (NIH Definition)
The failure to comply with applicable NIH Human Research Protections Program (HRPP) policies, IRB requirements, or regulatory requirements for the protection of human research subjects

Unanticipated Problem
Any incident, experience, or outcome that:

- Is unexpected in terms of nature, severity, or frequency in relation to

(a) the research risks that are described in the IRB-approved research protocol and informed consent document; Investigator's Brochure or other study documents, and
(b) the characteristics of the subject population being studied; A**ND**

- Is related or possibly related to participation in the research; **AND**
- Suggests that the research places subjects or others at a g*reater risk of harm* (including physical, psychological, economic, or social harm) than was previously known or recognized.

7.3.6.2 NIH IRB and Clinical Director Reporting Requirements

The Protocol PI will report in the NIH Problem Form to the NIH-IRB and NCI Clinical Director:

- All deaths, except deaths due to progressive disease
- All Protocol Deviations
- All Unanticipated Problems
- All non-compliance

Reports must be received within 7 days of PI awareness via iRIS.

7.3.6.3 NIH IRB Reporting of IND Safety Reports

Only IND Safety Reports that meet the definition an unanticipated problem will need to be reported to the NIH IRB.

7.4 Routine Adverse Event Reporting

All Adverse Events **must** be reported in routine study data submissions. **AEs reported through CTEP-AERS must <u>also</u> be reported in routine study data submissions**.

7.4.1 NIH IRB Requirements for PI Reporting at Continuing Review

Please use the following table for reporting adverse event at time of CR:

System Organ Class	CTCAE Term	Grade	# of events since last CR	Total # of events	Attribution to Research	Serious?	Unexpected?

The protocol PI will report to the NIH IRB:

1. A summary of all protocol deviations in a tabular format to include the date the deviation occurred, a brief description of the deviation and any corrective action.
2. A summary of any instances of non-compliance
3. A tabular summary of the following adverse events:

- All Grade 2 **unexpected** events that are possibly, probably or definitely related to the research;

- All Grade 3 and 4 events that are possibly, probably or definitely related to the research;
- All Grade 5 events regardless of attribution;
- All Serious Events regardless of attribution.

NOTE: Grade 1 events are not required to be reported.

7.5 Secondary Malignancy

A *secondary malignancy* is a cancer caused by treatment for a previous malignancy (e.g., treatment with investigational agent/intervention, radiation or chemotherapy). A secondary malignancy is not considered a metastasis of the initial neoplasm.

CTEP requires all secondary malignancies that occur following treatment with an agent under an NCI IND/IDE be reported via CTEP-AERS. Three options are available to describe the event:

- Leukemia secondary to oncology chemotherapy (e.g., acute myelocytic leukemia [AML])
- Myelodysplastic syndrome (MDS)
- Treatment-related secondary malignancy

Any malignancy possibly related to cancer treatment (including AML/MDS) should also be reported via the routine reporting mechanisms outlined in each protocol.

7.6 Second Malignancy

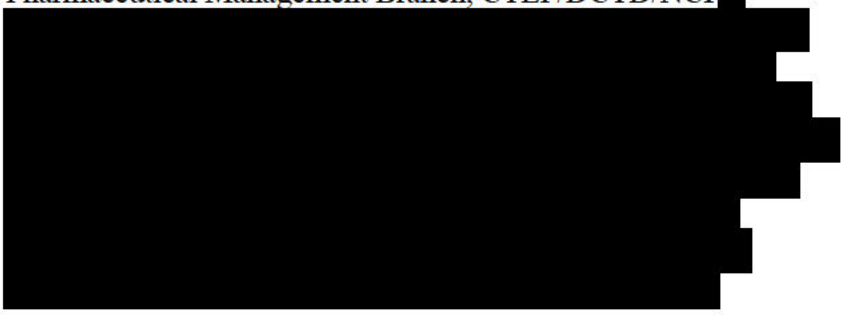
A second malignancy is one unrelated to the treatment of a prior malignancy (and is **NOT** a metastasis from the initial malignancy). Second malignancies require **ONLY** routine reporting via CDUS unless otherwise specified.

8 PHARMACEUTICAL INFORMATION

A list of the adverse events and potential risks associated with the investigational agent administered in this study can be found in Section 7.1.

8.1 AZD1775 (NSC 751084)

Chemical Name:	2-allyl-1-[6-(1-hydroxy-1-methylethyl) pyridin-2-yl]-6-{[4-(4-methylpiperazin-1-yl) phenyl]amino}-1,2-dihydro-3H-pyrazolo[3,4-d]pyrimidin-3-one hemihydrate
Other Names:	MK-1775, Adavosertib
Classification:	Inhibitor of Wee1-kinase
(CAS):	(1277170-60-1)
Molecular Formula:	$C_{27}H_{32}N_8O_2 \cdot 0.5H_2O$

Molecular Weight:	500.6
Physical Form:	AZD1775 is a crystalline, hemihydrate form of the drug substance.
Mode of Action:	AZD1775 is an inhibitor of Wee1-kinase. Wee1 is a tyrosine kinase upstream of CDC2 and is involved in regulation of cell cycle checkpoints, particularly the G2 checkpoint. As the majority of human cancers harbor abnormalities in the p53 pathway they become more dependent on S- and G2-phase checkpoints. In preclinical models, AZD1775 selectively enhanced chemotherapy-induced death of cells deficient in p53 signaling.
How Supplied:	AZD1775 is supplied by AstraZeneca and distributed by the Pharmaceutical Management Branch, CTEP/DCTD/NCI [redacted]  AZD1775 capsules may be repackaged from the manufacturer-supplied HDPE bottle into a pharmacy-supplied HDPE bottle for dispensing up to a four-week supply.
Storage:	The capsules are packaged in high density polyethylene (HDPE) bottles fitted with induction seals. Store at 2 to 30˚C (36 to 86˚F). Do not freeze. If a storage temperature excursion is identified, promptly return AZD1775 to between 2-30°C and quarantine the supplies. Provide a detailed report of the excursion (including documentation of temperature monitoring and duration of the excursion) to PMBAfterHours@mail.nih.gov for determination of suitability.
Stability:	Shelf-life surveillance studies of AZD1775 are ongoing.
Route of Administration:	Oral: Since the effect of food on AZD1775 has not been determined, patients should take AZD1775 either two hours before or two hours after a meal.
Potential Drug Interaction:	[redacted]

Availability

AZD1775 is an investigational agent supplied to investigators by the Division of Cancer Treatment and Diagnosis (DCTD), NCI. AZD1775 is provided to the NCI under a Collaborative Agreement between the Pharmaceutical Collaborator and the DCTD, NCI.

8.2 Agent Ordering, Accountability, and Investigator Brochure Availability

NCI-supplied agents may be requested by eligible participating Investigators (or their authorized designee) at each participating institution. The CTEP-assigned protocol number must be used for ordering all CTEP-supplied investigational agents. The eligible participating investigators at each participating institution must be registered with CTEP, DCTD through an annual submission of FDA Form 1572 (Statement of Investigator), NCI Biosketch, Agent Shipment Form, and Financial Disclosure Form (FDF). If there are several participating investigators at one institution, CTEP-supplied investigational agents for the study should be ordered under the name of one lead participating investigator at that institution.

In general, sites may order initial agent supplies when a subject is being screened for enrollment onto the study.

Active CTEP-registered investigators and investigator-designated shipping designees and ordering designees can submit agent requests through the PMB Online Agent Order Processing (OAOP) application. Access to OAOP requires the establishment of a CTEP Identity and Access Management (IAM) account and the maintenance of an "active" account status and a "current" password. For questions about drug orders, transfers, returns, or accountability, call or email PMB any time. Refer to the PMB's website for specific policies and guidelines related to agent management.

Agent Inventory Records - The investigator, or a responsible party designated by the investigator, must maintain a careful record of the receipt, dispensing and final disposition of agent received from the PMB using the NCI Investigational Agent (Drug) Accountability Record Form (DARF) for Oral Agents available on the CTEP forms page. Store and maintain separate NCI Investigational Agent Accountability Records for each agent, strength, formulation and ordering investigator on this protocol.

Investigator Brochure Availability - The current versions of the IBs for the agents will be accessible to site investigators and research staff through the PMB Online Agent Order Processing (OAOP) application. Access to OAOP requires the establishment of a CTEP Identity and Access Management (IAM) account and the maintenance of an "active" account status and a "current" password. Questions about IB access may be directed to the PMB IB coordinator via email.

Useful Links and Contacts

- CTEP Forms, Templates, Documents: http://ctep.cancer.gov/forms/
- NCI CTEP Investigator Registration: RCRHelpDesk@nih.gov
- PMB policies and guidelines: http://ctep.cancer.gov/branches/pmb/agent_management.htm
- PMB Online Agent Order Processing (OAOP) application: https://ctepcore.nci.nih.gov/OAOP
- CTEP Identity and Access Management (IAM) account: https://ctepcore.nci.nih.gov/iam/
- CTEP IAM account help: ctepreghelp@ctep.nci.nih.gov
- IB Coordinator: IBCoordinator@mail.nih.gov
- PMB email: PMBAfterHours@mail.nih.gov
- PMB phone and hours of service: (240) 276-6575 Monday through Friday between 8:30 am and 4:30 pm (ET)

9 CORRELATIVE/SPECIAL STUDIES

We plan to evaluate the in vivo molecular effects of AZD1775 using pre- and post-treatment tumor biopsy specimens and CTCs; blood will additionally be collected for PK analysis. Samples will be collected during cycle 1 at baseline and 2-5 hours after the 5th dose of AZD1775 or, as of Amendment M (11/20/16), prior to the 6th dose of AZD1775 on day 8 during cycle 1 only. In addition, an optional "restaging follow-up biopsy" may be performed on day 1 (± 2 days) of the cycle following any restaging at which a 10-20% increase in tumor volume is observed (according to RECIST criteria) if the patient has been on study for at least 4 cycles, or at time of disease progression. Patients have the option to undergo paired tumor biopsies as part of participation in this study during the escalation phase; paired tumor biopsies will be required during the expansion phase at the MTD (Expansion Arm A). Tumor biopsies will be optional in the expansion arm of BRCA patients. For Arm B on a once-daily dosing schedule, mandatory paired tumor biopsies will be pursued in up to 20 patients enrolled at the maximum tolerated daily dose to further evaluate PD endpoints.

Biopsy tissue quality will be monitored and accrual will stop once we have obtained 10 usable paired samples. Tumor biopsies will be collected for evaluation of levels of DNA-damage repair and apoptosis after the 5th dose of AZD1775 during cycle 1.

9.1 Pharmacokinetics

AZD1775 will be assayed in plasma from all patients. Blood samples for PK analyses will be collected during cycle 1 only. Blood samples (6 mL) will be collected in lavender-top (EDTA) tubes and will be stored at -20°C until all time points (see Section 9.1.1) have been collected for that cycle. Samples will be shipped to Covance (See Section 9.1.2).

9.1.1 Blood Samples

Blood samples for PK analyses on **Arm A** will be collected at the following time points:
Cycle 1, Day 1: prior to treatment and at 1, 2, 4, 6, and 8 hours after the first dose
Cycle 1, Day 2: prior to the 3rd dose
Cycle 1, Day 3: prior to the 5th dose, and at 1, 2, 4, 6, and 8 hours after the 5th dose.
Additional PK sampling will be optional (but encouraged). This sampling would occur 24 and 48 hours after the 5th dose of AZD1775, and will occur on Days 4 and 5, respectively.

Blood samples for PK analyses on **Arm B** will be collected at the following time points:
Cycle 1, Day 1: prior to treatment and at 2 and 4 hours after the first dose
Cycle 1, Day 8: prior to the eighth dose

9.1.2 Laboratory Contact

PK plasma samples will be shipped in 10 kg of dry ice to:
Covance Central Laboratory Services
Attn: Phyllis Sellars – Special Handling
8211 SciCor Drive
Indianapolis, Indiana 46214
Phone: 317-271-1200

9.2 Pharmacodynamics

Evaluation of drug effect on DNA damage response will be performed by immunofluorescence assays for measurement of DNA damage-repair markers, such as Rad51, pNbs1, and γH2AX. Evaluation of caspase-3 by immunofluorescence assays will be performed on biopsy specimens as a marker of drug effect on apoptosis. Evaluation of Wee1 kinase inhibition will be performed by an immunofluorescence assay for pY15-

Cdk.

For the twice-daily dosing schedule (Arm A), CTCs will be obtained in all patients pre-dose, on C1D3 and C2D3 after the 5th administration of AZD1775, for evaluation of γH2AX as a marker of drug effect on apoptosis. For the once-daily dosing schedule (Arm B), CTCs will be obtained in all patients on day 1 (prior to AZD1775 administration), on day 4 (4 ± 1 hours after AZD1775 administration), on day 8 prior to the dose of AZD1775, on day 8 (4 ± 1 hours after AZD1775 administration), and on day 11 (4 ± 1 hours after AZD1775 administration). Additional optional CTC samples will be obtained at each restaging and at time of disease progression or "restaging follow-up" biopsy. Arm B CTC specimens will be evaluated for γH2AX, pHH3, pNbs1, and pY15-Cdk.

9.2.1 Laboratory Contact

At least 24 hours prior to tumor biopsy or blood sample collection, the research nurse will contact the NCI Phase I/II PK/PD Support Group in NIH Building 10: E-mail (preferred): NCIPK-PDsupportgroup@mail.nih.gov, Pager (preferred): 102-12798, Phone: 240-858-3963, Fax: 301-480-5871. For biopsies, tubes pre-labeled with the information specified in Section 9.2.4, biopsy date, and site of tissue biopsy will be provided. Initial processing and shipping of the samples will be completed as described below.

9.2.2 Blood Collection for CTC Studies (optional)*

Whole blood will be collected aseptically by venipuncture or from a venous port into one 4-mL sodium heparin green glass tube (Arm A) or two 4-mL EDTA tubes (Arm B); the collected blood samples should be shipped to PADIS on the day of sample collection to avoid excessive CTC cell death prior to specimen processing. Blood samples for CTCs will be collected from patients enrolled on Arm A at the following time points:

- Cycle 1 prior to treatment,
- Cycle 1, day 3: 2-5 hours after the 5th dose
- Cycle 2, day 3: 2-5 hours after the 5th dose

For patients enrolled on Arm B, on the once daily dosing schedule, blood samples for CTCs (two 4-mL EDTA tubes per time point) will be collected from all patients at the following time points during each cycle:

- Day 1, prior to treatment,
- Day 4, 4 ± 1 hours after AZD1775 administration
- Day 8, prior to treatment
- Day 8, 4 ± 1 hours after AZD1775 administration
- Day 11, 4 ± 1 hours after AZD1775 administration
- Every 2 cycles at restaging (3 or 4 cycles for patients on study more than one

or three years, respectively)
- Optionally, at time of disease progression or "restaging follow-up biopsy," on day 1 (± 2 days) of the cycle following any restaging at which a 10-20% increase in tumor volume is observed (according to RECIST criteria) if the patient has been on study for at least 4 cycles.

9.2.3 Blood Collection for ImmunoPD Studies (PBMCs and serum; optional) for the Laboratory of Tumor Immunology

For patients enrolled on Arm B once-daily dosing schedule, optional blood samples for immunoPD will be collected into one 6-mL sodium heparin tube and one 7-mL SST at the following time points:
- Cycle 1 prior to treatment,
- Cycle 1, day 8 prior to treatment (+/- 1 day for scheduling)

Sample processing will be performed by the Clinical Services Program (CSP), FNLCR; flow cytometry will be performed by LTIB.

Samples should be kept at room temperature prior to processing. Shipment by CSP Courier may be arranged by contacting Jennifer Bangh, FNLCR, Tel.: 301-846-5893.

9.2.4 Tumor Biopsies

9.2.4.1 Timing of tumor biopsies

Biopsies will be optional during the escalation phase and mandatory in the 6 additional patients enrolled at the MTD during the expansion phase (Expansion Arm A). Tumor biopsies will remain optional in the expansion arm of BRCA patients and follow the biopsy collection times as follows. Biopsies will be collected:
- before drug administration on study (baseline), and
- on cycle 1, day 3; 2-5 hours after the 5th dose of AZD1775

For the patients enrolled on Arm B, biopsies will remain optional during the escalation phase, but will be mandatory in up to 20 patients enrolled at the maximum tolerated single daily dosing schedule during the expansion phase. Biopsies will be collected:
- before drug administration on study (baseline), and
- on cycle 1, day 8; prior to the 6th dose of AZD1775 (+/- 1 day)
- optionally, at disease progression or on day 1 (± 2 days) of the cycle following any restaging at which a 10-20% increase in tumor volume is observed (according to RECIST criteria) if the patient has been on study for at least 4 cycles. (For example, a biopsy would be performed on cycle 6 day 1 for a 10-20% tumor volume increase detected at the restaging following cycle 4).

9.2.4.2 Biopsy Procedure

Serial tumor biopsies will be obtained by the Interventional Radiology team by a percutaneous approach. It is preferred that up to 5 core biopsies ≥18 gauge in diameter and ≥1 cm in length will be obtained during each procedure if considered safe and feasible. It is estimated that there will be between 2 million–5 million cells from each biopsy. If a site is deemed appropriate for biopsy with minimal risk to the participant by agreement between the investigators and Interventional Radiology team, an attempt for biopsy will be made.

The use of imaging to facilitate biopsies will be decided by members of the Interventional Radiology team and may include ultrasound, CT scan, or MRI. Should a CT scan be needed for biopsy, the number of scans for each procedure will be limited to the minimum number needed to safely obtain a biopsy. Tumor biopsies and local anesthesia will be administered only if they are considered to be of low risk to the participant, as determined by the investigators and Interventional Radiology.

Acceptable biopsy procedures are: percutaneous biopsy with local anesthetic; excisional cutaneous biopsy with local anesthetic; other biopsy with local anesthetic and/or sedation that has been shown to have a risk of severe complications < 2%; biopsy with removal of additional tumor tissue during a medically necessary mediastinoscopy, laparoscopy, gastrointestinal endoscopy, bronchoscopy, or craniotomy; removal of additional tumor tissue during a medically necessary surgical procedure. No open surgical, laparoscopic endoscopic procedure will be performed solely to obtain a biopsy for this protocol.

If the participant chooses not to undergo tumor biopsy, he/she will still remain in the study and receive study medication, and all the other correlative studies will be performed.

Tumor biopsies are optional during the escalation phase and mandatory for the additional patients enrolled at the MTD during the BRCA-arm expansion phase. Baseline biopsies will be performed following patient enrolling on study. For Arm B, if the patient had a biopsy within the past 3 months and had no treatment since that biopsy, an attempt will be made to obtain and process that tissue as baseline biopsy. If the tissue is not of sufficient quality or cannot be used for protocol-specified PD studies, a percutaneous biopsy will be performed. If an initial attempt at percutaneous biopsy is unsuccessful, the patient will be given an option to proceed with a repeated attempt at percutaneous biopsy. A separate consent form must be signed for each biopsy procedure, so patients may choose not to undergo subsequent biopsies. If the baseline biopsy is unsuccessful or the patient refuses to undergo subsequent biopsies, no further biopsies will be performed but the patient will remain on study, receive study medication, and other correlative studies will be performed.

9.2.4.3 Solid Tumor Biopsy Processing

Up to 5 tissue cores will be collected. Cores will be flash frozen in liquid nitrogen and submitted to Dr. Kinders' laboratory for evaluation of DNA damage markers, and H&E pathology evaluation. The frozen biopsy specimens are transferred to PADIS, where the core biopsy samples are stored at -80°C and subsequently processed within 7-10 days for analysis. Biopsy samples will be analyzed for γH2AX and pTyr15-Cdk. Additional analyses of markers of DNA damage will depend on tissue availability. One half-core pre-treatment flash frozen biopsy will be saved and stored for future analyses: targeted sequencing for DNA repair defects and p53 mutational analysis is being discussed with the laboratories. Such studies, if performed, will be conducted following an amendment to the current protocol.

Biopsies for PD analysis will be shipped on dry ice to:

Attention: Dan Danner
NCI-F/FNLCR
1073 Beasley Street, Building 1073
Fort Detrick
Frederick, MD 21701
Phone: 301-846-5748
NCI_PD_Support@mail.nih.gov

Blood for CTC analysis will be shipped at room temperature on the day of sample collection to:
Attention: Dan Danner
NCI-F/FNLCR
1073 Beasley Street, Building 1073
Fort Detrick
Frederick, MD 21701
Phone: 301-846-5748
NCI_PD_Support_CellSearch@mail.nih.gov

Shipment should be by CSP Courier and may be arranged by contacting Jennifer Bangh, FNLCR, Tel.: 301-846-5893

9.2.5 Sample Collection and Processing

Biospecimens will be collected and processed using validated SOPs that will ensure both specimen quality and patient confidentiality pursuant to informed consent provisions. Information about each specimen (e.g., blood, tumor biopsy, circulating tumor cells, per specific protocol) will be recorded on a PK/PD collection worksheet included in Appendix F.

Using a computerized inventory system and a backup hardcopy process, all specimen collection and processing steps will be documented and the specific

location of each specimen will be tracked. Each new specimen collected will be assigned a unique barcode identifier that can be linked to the original specimen collected and other relevant information within the inventory system. To ensure patient confidentiality, only containers used for the initial specimen collections will be labeled with patient identifiers.

Only the barcode identifier will be applied to all subsequent specimen containers. When specimens are processed and aliquoted, no patient information will be included on the new containers. Original specimen containers will be discarded. Only barcode-labeled specimens without patient identifiers will be shipped for analysis and/or storage. Specimen labels will indicate: CTEP protocol number, unique patient accession number, 3-digit sample number (see list below), collection time, and total volume collected, as appropriate. Samples from sets of at least three patients will be grouped for scientific analysis.

Standardized 3-digit sample collection numbers:
100 series: urine for PK
200 series: blood for PK
300 series: blood for PD
400 series: blood for circulating tumor cells (CTCs)
500 series: tumor biopsies
800 series: blood for pharmacogenomics or other research use

The inventory process contains other security provisions sufficient to safeguard patient privacy and confidentiality. Access to the inventory system and associated documents will be restricted to appropriate individuals. Requests to use specimens stored in the repository must be approved. The only patient information available in the inventory system will be the patient sex, diagnosis, and level of informed consent given. SOPs ensure that any changes in informed consent made by a patient and relayed to the PI will be reflected in the inventory system to ensure that specimens are destroyed as appropriate. All laboratory personnel will be trained to adhere to SOPs and will be monitored for high-quality performance.

Any pharmacokinetic sample remaining after analysis for AZD1775 may be used for exploratory DDI, metabolite, or biomarker analyses. For example, circulating citrulline concentration is emerging as a biomarker candidate for the assessment of intestinal function and chemotherapy-induced gastrointestinal toxicity, and preclinical studies of AZD1775 found a reasonable correlation between villus atrophy and decreases in body weight and plasma citrulline. Thus, remaining plasma PK samples may be used to evaluate changes in plasma citrulline and its relationship to GI toxicities.

Any new use of these samples will require prospective IRB review and approval. Access to these samples will only be granted following IRB approval of an additional protocol, granting the rights to use the material.

If at any time, a patient withdraws from the study and does not wish for their existing samples to be utilized, the individual must provide a written request. Following receipt of this request, the samples will be destroyed (or returned to the patient, if so requested), and reported as such to the IRB. Any samples lost (in transit or by a researcher) or destroyed due to unknown sample integrity (i.e., broken freezer allows for extensive sample thawing, etc.) will be reported as such to the IRB.

9.2.5.1 Human Data Sharing Plan

What data will be shared?

We will share human data generated in this research for future research as follows:

x De-identified data in an NIH-funded or approved public repository
x Identified data in BTRIS (automatic for activities in the Clinical Center)
x De-identified or identified data with approved outside collaborators under appropriate agreements

How and where will the data be shared?

Data will be shared through:

x An NIH-funded or approved public repository: clinicaltrials.gov
x BTRIS (automatic for activities in the Clinical Center)
x Approved outside collaborators under appropriate individual agreements
x Publication and/or public presentations

When will the data be shared?

x At the time of publication or shortly thereafter

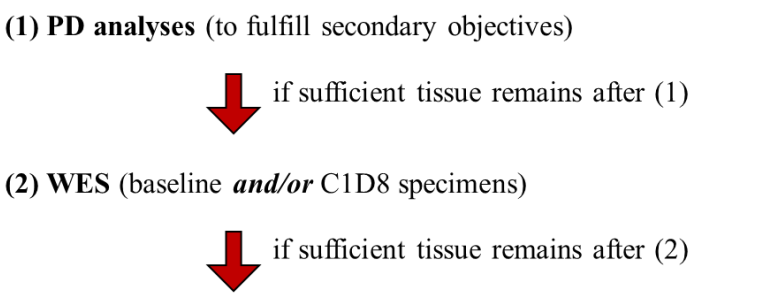
9.3 Exploratory whole-exome and RNA sequencing

For active Expansion Arm B patients enrolled at the time of Amendment P (11/2/17) approval or thereafter, baseline, cycle 1 day 8, and progression/"restaging follow-up" tumor biopsy tissue remaining after completion of PD biomarker analyses will be used for exploratory whole-exome sequencing (WES) and RNA sequencing (RNA-seq) as appropriate to identify tumor genomic alterations and gene expression patterns potentially associated with AZD1775 antitumor activity, including loss of function mutations in SETD2[30] and amplification or overexpression of CCNE1 or MYC[29]. Baseline and cycle 1 day 8 blood samples will also be collected for sequencing to enable comparison of tumor and germline sequences. Tissue specimens used for sequencing analyses will be de-identified, with only limited clinical annotation (e.g., response to AZD1775) maintained for each specimen. Active Expansion Arm B patients already enrolled at the time of Amendment P approval will be re-consented to the new, "genetic analysis" consent form prior to use of their specimens for exploratory sequencing, and, if both pre- and post- biopsies have

already been collected from such patients, a blood sample will be collected at time of re-consent for germline DNA analysis, as described below.

Biopsy cores and tissue sections are to be used for exploratory WES or RNA-seq only after PD biomarker analyses have been completed. Tissue availability will be tracked in real time using Labmatrix. Although the MoCha Laboratory is CLIA-certified, these sequencing studies will not be done per CLIA specifications as the data are not returned to the patients or used for clinical decision making. WES analysis will be prioritized over RNA-seq (see "Biopsy specimen use prioritization" diagram, below). WES and RNA-seq analysis of both baseline and cycle 1 day 8 tumor tissue specimens is preferred; however, WES may be performed for a sample from only one time point depending on tissue availability. RNA-seq may be performed for baseline specimens only if no cycle 1 day 8 tissue is available.

Biopsy specimen use prioritization



9.3.1 Tumor samples

Biopsy tissue suitable for exploratory sequencing analyses includes frozen or FFPE tumor biopsy cores (< 1 core per sequencing run) or a series of slide-mounted FFPE tumor tissue sections (approximately 20 4 μm-thick tumor sections per sequencing run).

For all patients, frozen (cores) or FFPE (cores or slides) tumor tissue will be submitted for genetic analyses to the MoCha Laboratory. Samples labeled with unique patient IDs only will be transferred directly from PADIS to Dr. Mickey Williams' laboratory. **Do NOT include patient identifiers (e.g., medical record number, patient name, or initials) with the samples.**

MoCha Lab, Frederick National Laboratory for Cancer Research
Fort Detrick
Building 433 Room #3/#15
Frederick, MD 21702
NCIFrederickMoChaRND@mail.nih.gov

The biopsy specimen will be extracted for nucleic acids using the Qiagen FFPET All-Prep procedure. DNA and RNA will be assessed for quantity and quality by spectroscopy (OD 260/280) and a PCR-based amplification quality assessment test. All specimens that meet necessary quantity and quality criteria will be used for whole-exome and RNA sequencing as appropriate.

9.3.2 Blood samples

Optional whole blood samples will be collected at baseline and cycle 1 day 8 to obtain mononuclear cells. The nucleic acid extracted from the patient's mononuclear cells will be sequenced to compare somatic mutational status to the tumor biopsy specimen. For active Expansion Arm B patients already enrolled at the time of Amendment P approval who are re-consented to the new, "genetic analysis" consent form after both pre- and post- biopsies have already been collected, an optional blood sample will be collected at time of re-consent.

One 7-mL K3 EDTA tube should be collected on the same day as the biopsy (either before or after the biopsy procedure) and shipped at ambient temperature. Samples should be labeled with only the unique patient ID. **Do NOT include patient identifiers (e.g., medical record number, patient name, or initials) with the samples.**

Shipment of blood samples should be by CSP Courier and may be arranged by contacting Jenn Bangh, FNLCR, Tel.: 301-846-5893. Samples should be sent to:

MoCha Lab, Frederick National Laboratory for Cancer Research
Fort Detrick
Building 433 Room #3/#15
Frederick, MD 21702
NCIFrederickMoChaRND@mail.nih.gov

10 STUDY CALENDAR

Eligibility screening evaluations are to be performed within 8 days prior to enrollment, with the exception of tumor imaging scans which must be performed within 28 days prior to enrollment (see Section 3.4). Baseline history, physical examination, laboratory evaluations, urinalysis, and ECG are to be conducted within 8 days prior to start of protocol therapy. If protocol therapy is started within 8 days of the screening evaluations, the screening evaluations may be used as baseline measurements. If >8 days have passed since the screening evaluations, the medical history, physical examination, laboratory evaluations, urinalysis, and ECG must be repeated prior to starting protocol therapy. Scans/x-rays and ECHO must be done within 28 days prior to the start of protocol therapy. In the event that the patient's condition is deteriorating, laboratory evaluations should be repeated within 48 hours prior to initiation of the next cycle of therapy. Start of next cycle may be delayed for up to 1 week to accommodate scheduling conflicts. For cycle 2 and beyond, history, physical

examination, and laboratory evaluations can be performed up to 3 days before the start of the next cycle.

	Pre-Study Eligibility Screening	Baseline Clinical Evaluation	C1 W 1	C1 W 2	C1 W 3	C2 W 1	C2 W 2	C2 W 3	C3 W 1	C3 W 2	C3 W 3	C4 W 1	C4 W 2	C4 W 3	Off Treatment[k]
AZD1775 DL1[a]			X			X			X			X			
AZD1775 DL2 on[a]			X	X		X	X		X	X		X	X		
Informed consent	X														
Demographics	X														
Medical history	X	X[l]		X		X			X			X			
Concurrent meds	X	X[l]	X----------											----------X	
Physical exam[b]	X	X[l]		X		X			X			X			X
Vital signs	X	X[l]		X		X			X			X			X
Height	X	X[l]													
Weight	X	X[l]				X			X			X			X
Performance status	X	X[l]				X			X			X			X
CBC w/diff, plts[c]	X	X[l]		X	X	X			X			X			X
Serum chemistry[c]	X	X[l]		X	X	X			X			X			X
PT, INR, PTT[d]	X	X[l]													
β-HCG[e]	X					X			X			X			
Adverse event evaluation			X----------											----------X	X
Radiologic evaluation	X	X[l]	Radiologic measurements should be performed every 2 cycles (see Section 11.2 for patients on study for more than one year). Documentation (radiologic) must be provided for patients removed from study for progressive disease.												X
Tumor biopsy[f]			Biopsies will be performed as described in Section 9.2.4.												
PK blood sampling[g]			X	X											
Circulating tumor cells[h]			Blood for circulating tumor cells (optional) will be collected at multiple time points as specified in Section 9.2.2												
Immunoblood sampling[i]			X	X											
ECG[j]	X	X[l]													

a: **For dose level 1,** AZD1775 will be administered orally for 5 doses each cycle, with doses administered approximately 12 hours apart starting with the first dose for that week (for example, if the patient takes the first dose at 9 AM on Monday then the next dose will be administered at 9 pm that evening). **Starting at dose level 2 and onwards**, AZD1775 will be administered orally for 5 doses for the first 2 weeks of each cycle, with doses administered approximately 12 hours apart starting with the first dose for that week (as explained above except it will occur over the first two weeks of the cycle). Each cycle is 21 days (± 1 day for scheduling). For Arm B, AZD1775 will be administered orally once daily, for 5 doses each week, for two consecutive weeks. Cycle length will remain 21 days (± 1 day for scheduling).

b: Physical examination at the Clinical Center should be performed at the start of each cycle (up to 3 days before start of a new cycle)
c: Serum chemistry (albumin, total bilirubin, calcium, creatinine, phosphorus, magnesium, potassium, sodium, SGOT [AST], SGPT [ALT]); CBC w/diff, platelets weekly during cycle 1, then every 3 weeks at the start of each subsequent cycle (up to 3 days before start of new cycle). If clinically indicated, labs may be obtained with more frequency with subsequent cycles.
d. PT/INR, PTT prior to biopsy.
e: Serum or urine pregnancy test (women of childbearing potential) within 1 week prior to enrollment, prior to starting each treatment cycle, and as clinically indicated.
f. Tumor biopsies will be performed as specified in Section 9.2.4. Tumor biopsies are optional during the escalation phase, but will be mandatory in the 6 patients enrolled at MTD (Expansion Arm A) for evaluation of PK and PD endpoints. Tumor biopsies will be optional in the 6 patients in the BRCA-1 or -2 mutation expansion arm. For patients enrolled on Arm B at once daily dosing, tumor biopsies will remain optional until the maximum tolerated daily dosing is established, at which point up to 20 additional patients will be enrolled and tumor biopsies will be mandatory. With Amendment P (11/2/17), on the day of each biopsy procedure (either before or after the biopsy procedure), an optional blood sample should be collected into a 7-mL K3 EDTA tube for exploratory sequencing analyses (see Section 9.3.2).
g: PK samples for Arm A will be performed at the following time-points:
C1D1: Pre-dose, at 1, 2, 4, 6, and 8 hours after the first dose
C1D2: Prior to the 3rd dose
C1D3: Prior to the 5th dose, and 1, 2, 4, 6, 8 hours after the 5th dose.
Additional PK sampling will be optional (but encouraged). This sampling would occur 24 and 48 hours after the 5th dose of AZD1775, and will occur on Days 4 and 5, respectively.
For patients enrolled on Arm B, PK samples will be performed at the following time-points:
C1D1: Prior to the first dose and at 2 and 4 hr after the first dose,
C1D8: Prior to the 6^{th} dose
h: Circulating tumor cells (optional) will be collected at multiple time points as specified in Section 9.2.2.
i: Optional blood and serum for immune PD studies (Gulley Lab) will be collected for Arm B patients prior to treatment and prior to dosing on cycle 1 day 8 (+/- 1 day).
j: ECG will be performed at baseline for evaluation of QTc and as clinically indicated thereafter.
k: Off-study evaluation.
l: Values from eligibility screening tests may be used as baseline evaluation values if the test was performed within 8 days of start of protocol therapy (or, for radiologic evaluation and tumor measurement, within 28 days of start of protocol therapy). See Section 3.4 and Section 5.

11 MEASUREMENT OF EFFECT

11.1 Antitumor Effect – Solid Tumors

Although response is not the primary endpoint of this trial, patients with measurable disease will be assessed by standard criteria. For the purposes of this study, patients should be re-evaluated for response every 6 weeks (every 2 cycles; 3 or 4 cycles for patients on study more than one or three years, respectively). In addition to a baseline scan, confirmatory scans should also be obtained at least 4 weeks following initial documentation of objective response.

Response and progression will be evaluated in this study using the new international criteria proposed by the revised Response Evaluation Criteria in Solid Tumors (RECIST) guideline (version 1.1) [*Eur J Ca* 45:228-247, 2009]. Changes in the largest diameter (unidimensional measurement) of the tumor lesions and the shortest diameter in the case of malignant lymph nodes are used in the RECIST criteria.

11.1.1 Definitions

Evaluable for toxicity: All patients will be evaluable for toxicity from the time of their first treatment with AZD1775.

Evaluable for objective response: Only those patients who have measurable disease present at baseline, have received at least one cycle of therapy, and have had their disease re-evaluated will be considered evaluable for response. These patients will have their response classified according to the definitions stated below. (Note: Patients who exhibit objective disease progression prior to the end of cycle 1 will also be considered evaluable.)

Evaluable Non-Target Disease Response: Patients who have lesions present at baseline that are evaluable but do not meet the definitions of measurable disease, have received at least one cycle of therapy, and have had their disease re-evaluated will be considered evaluable for non-target disease. The response assessment is based on the presence, absence, or unequivocal progression of the lesions.

11.1.2 Disease Parameters

Measurable disease: Measurable lesions are defined as those that can be accurately measured in at least one dimension (longest diameter to be recorded) as ≥20 mm by chest x-ray or as ≥10 mm with CT scan, MRI, or calipers by clinical exam. All tumor measurements must be recorded in millimeters (or decimal fractions of centimeters).

Malignant lymph nodes: To be considered pathologically enlarged and measurable, a lymph node must be ≥15 mm in short axis when assessed by CT scan (CT scan slice thickness recommended to be no greater than 5 mm). At baseline and in follow-up, only the short axis will be measured and followed.

Non-measurable disease: All other lesions (or sites of disease), including small lesions (longest diameter <10 mm or pathological lymph nodes with ≥10 to <15 mm short axis), are considered non-measurable disease. Bone lesions, leptomeningeal disease, ascites, pleural/pericardial effusions, lymphangitis cutis/pulmonitis, inflammatory breast disease, and abdominal masses (not followed by CT or MRI), are considered as non-measurable.

Note: Cystic lesions that meet the criteria for radiographically defined simple cysts should not be considered as malignant lesions (neither measurable nor non-measurable) since they are, by definition, simple cysts.

'Cystic lesions' thought to represent cystic metastases can be considered as measurable lesions, if they meet the definition of measurability described above. However, if non-cystic lesions are present in the same patient, these are preferred for selection as target lesions.

Target lesions: All measurable lesions up to a maximum of 2 lesions per organ and 5 lesions in total, representative of all involved organs, should be identified as **target lesions** and recorded and measured at baseline. Target lesions should be selected on the basis of their size (lesions with the longest diameter), be representative of all involved organs, but in addition should be those that lend themselves to reproducible repeated measurements. It may be the case that, on occasion, the largest lesion does not lend itself to reproducible measurement in which circumstance the next largest lesion which can be measured reproducibly should be selected. A sum of the diameters (longest for non-nodal lesions, short axis for nodal lesions) for all target lesions will be calculated and reported as the baseline sum diameters. If lymph nodes are to be included in the sum, then only the short axis is added into the sum. The baseline sum diameters will be used as reference to further characterize any objective tumor regression in the measurable dimension of the disease.

Non-target lesions: All other lesions (or sites of disease) including any measurable lesions over and above the 5 target lesions should be identified as **non-target lesions** and should also be recorded at baseline. Measurements of these lesions are not required, but the presence, absence, or in rare cases unequivocal progression of each should be noted throughout follow-up.

11.2 Guidelines for Evaluation of Measurable Disease

All measurements should be taken and recorded in metric notation using a ruler or calipers. All baseline evaluations should be performed as closely as possible to the beginning of treatment and never more than 4 weeks before the beginning of the treatment.

The same method of assessment and the same technique should be used to characterize each identified and reported lesion at baseline and during follow-up. Imaging-based evaluation is preferred to evaluation by clinical examination unless the lesion(s) being followed cannot be imaged but are assessable by clinical exam.

Clinical lesions: Clinical lesions will only be considered measurable when they are superficial (e.g., skin nodules and palpable lymph nodes) and ≥10 mm diameter as assessed using calipers (e.g., skin nodules). In the case of skin lesions, documentation by color photography, including a ruler to estimate the size of the lesion, is recommended.

Chest x-ray: Lesions on chest x-ray are acceptable as measurable lesions when they are clearly defined and surrounded by aerated lung. However, CT is preferable.

Conventional CT and MRI: This guideline has defined measurability of lesions on CT scan based on the assumption that CT slice thickness is 5 mm or less. If CT scans have slice thickness greater than 5 mm, the minimum size for a measurable lesion should be twice the slice thickness. MRI is also acceptable in certain situations (e.g. for body scans).

As with CT, if an MRI is performed, the technical specifications of the scanning sequences used should be optimized for the evaluation of the type and site of disease. Furthermore, as with CT, the modality used at follow-up should be the same as was used at baseline and the lesions should be measured/assessed on the same pulse sequence. It is beyond the scope of the RECIST guidelines to prescribe specific MRI pulse sequence parameters for all scanners, body parts, and diseases. Ideally, the same type of scanner should be used and the image acquisition protocol should be followed as closely as possible to prior scans. Body scans should be performed with breath-hold scanning techniques, if possible.

PET-CT: At present, the low dose or attenuation correction CT portion of a combined PET-CT is not always of optimal diagnostic CT quality for use with RECIST measurements. However, if the site can document that the CT performed as part of a PET-CT is of identical diagnostic quality to a diagnostic CT (with IV and oral contrast), then the CT portion of the PET-CT can be used for RECIST measurements and can be used interchangeably with conventional CT in accurately measuring cancer lesions over time. Note, however, that the PET portion of the CT introduces additional data which may bias an investigator if it is not routinely or serially performed.

Ultrasound: Ultrasound is not useful in assessment of lesion size and should not be used as a method of measurement. Ultrasound examinations cannot be reproduced in their entirety for independent review at a later date and, because they are operator dependent, it cannot be guaranteed that the same technique and measurements will be taken from one assessment to the next. If new lesions are identified by ultrasound in the course of the study, confirmation by CT or MRI is advised. If there is concern about radiation exposure at CT, MRI may be used instead of CT in selected instances.

Endoscopy, Laparoscopy: The utilization of these techniques for objective tumor evaluation is not advised. However, such techniques may be useful to confirm complete pathological response when biopsies are obtained or to determine relapse in trials where recurrence following complete response (CR) or surgical resection is an endpoint.

Tumor markers: Tumor markers alone cannot be used to assess response. If markers are initially above the upper normal limit, they must normalize for a patient to be considered in complete clinical response. Specific guidelines for both CA-125 response (in recurrent ovarian cancer) and PSA response (in recurrent prostate cancer) have been published

[*JNCI* 96:487-488, 2004; *J Clin Oncol* 17, 3461-3467, 1999; *J Clin Oncol* 26:1148-1159, 2008]. In addition, the Gynecologic Cancer Intergroup has developed CA-125 progression criteria which are to be integrated with objective tumor assessment for use in first-line trials in ovarian cancer [*JNCI* 92:1534-1535, 2000].

Cytology, Histology: These techniques can be used to differentiate between partial responses (PR) and complete responses (CR) in rare cases (*e.g.*, residual lesions in tumor types, such as germ cell tumors, where known residual benign tumors can remain).

The cytological confirmation of the neoplastic origin of any effusion that appears or worsens during treatment when the measurable tumor has met criteria for response or stable disease is mandatory to differentiate between response or stable disease (an effusion may be a side effect of the treatment) and progressive disease.

FDG-PET: While FDG-PET response assessments need additional study, it is sometimes reasonable to incorporate the use of FDG-PET scanning to complement CT scanning in assessment of progression (particularly possible 'new' disease). New lesions on the basis of FDG-PET imaging can be identified according to the following algorithm:

a. Negative FDG-PET at baseline, with a positive FDG-PET at follow-up is a sign of PD based on a new lesion.
b. No FDG-PET at baseline and a positive FDG-PET at follow-up: If the positive FDG-PET at follow-up corresponds to a new site of disease confirmed by CT, this is PD. If the positive FDG-PET at follow-up is not confirmed as a new site of disease on CT, additional follow-up CT scans are needed to determine if there is truly progression occurring at that site (if so, the date of PD will be the date of the initial abnormal FDG-PET scan). If the positive FDG-PET at follow-up corresponds to a pre-existing site of disease on CT that is not progressing on the basis of the anatomic images, this is not PD.
c. FDG-PET may be used to upgrade a response to a CR in a manner similar to a biopsy in cases where a residual radiographic abnormality is thought to represent fibrosis or scarring. The use of FDG-PET in this circumstance should be prospectively described in the protocol and supported by disease-specific medical literature for the indication. However, it must be acknowledged that both approaches may lead to false positive CR due to limitations of FDG-PET and biopsy resolution/sensitivity.

Note: A 'positive' FDG-PET scan lesion means one which is FDG avid with an uptake greater than twice that of the surrounding tissue on the attenuation corrected image.

11.3 Response Criteria

11.3.1 Evaluation of Target Lesions

Complete Response (CR):	Disappearance of all target lesions. Any pathological lymph nodes (whether target or non-target) must have reduction in short axis to <10 mm.

Partial Response (PR):	At least a 30% decrease in the sum of the diameters of target lesions, taking as reference the baseline sum of diameters.
Progressive Disease (PD):	At least a 20% increase in the sum of the diameters of target lesions, taking as reference the smallest sum on study (this includes the baseline sum if that is the smallest on study). In addition to the relative increase of 20%, the sum must also demonstrate an absolute increase of at least 5 mm. (Note: the appearance of one or more new lesions is also considered progressions).
Stable Disease (SD):	Neither sufficient shrinkage to qualify for PR nor sufficient increase to qualify for PD, taking as reference the smallest sum of diameters while on study.

11.3.2 Evaluation of Non-Target Lesions

Complete Response (CR):	Disappearance of all non-target lesions and normalization of tumor marker level. All lymph nodes must be non-pathological in size (<10 mm short axis). Note: If tumor markers are initially above the upper normal limit, they must normalize for a patient to be considered in complete clinical response.
Non-CR/Non-PD:	Persistence of one or more non-target lesion(s) and/or maintenance of tumor marker level above the normal limits.
Progressive Disease (PD):	Appearance of one or more new lesions and/or *unequivocal progression* of existing non-target lesions. *Unequivocal progression* should not normally trump target lesion status. It must be representative of overall disease status change, not a single lesion increase.

Although a clear progression of "non-target" lesions only is exceptional, the opinion of the treating physician should prevail in such circumstances, and the progression status should be confirmed at a later time by the review panel (or Principal Investigator).

11.3.3 Evaluation of Best Overall Response

The best overall response is the best response recorded from the start of the treatment until disease progression/recurrence (taking as reference for progressive disease the smallest measurements recorded since the treatment started). The patient's best response assignment will depend on the achievement of both measurement and confirmation criteria.

For Patients with Measurable Disease (i.e., Target Disease)

Target Lesions	Non-Target Lesions	New Lesions	Overall Response	Best Overall Response when Confirmation is Required*
CR	CR	No	CR	≥4 wks. Confirmation**
CR	Non-CR/Non-PD	No	PR	≥4 wks. Confirmation**
CR	Not evaluated	No	PR	
PR	Non-CR/Non-PD/not evaluated	No	PR	
SD	Non-CR/Non-PD/not evaluated	No	SD	Documented at least once ≥4 wks. from baseline**
PD	Any	Yes or No	PD	no prior SD, PR or CR
Any	PD***	Yes or No	PD	
Any	Any	Yes	PD	

* See RECIST 1.1 manuscript for further details on what is evidence of a new lesion.
** Only for non-randomized trials with response as primary endpoint.
*** In exceptional circumstances, unequivocal progression in non-target lesions may be accepted as disease progression.

Note: Patients with a global deterioration of health status requiring discontinuation of treatment without objective evidence of disease progression at that time should be reported as "*symptomatic deterioration.*" Every effort should be made to document the objective progression even after discontinuation of treatment.

For Patients with Non-Measurable Disease (i.e., Non-Target Disease)

Non-Target Lesions	New Lesions	Overall Response
CR	No	CR
Non-CR/non-PD	No	Non-CR/non-PD*
Not all evaluated	No	not evaluated
Unequivocal PD	Yes or No	PD
Any	Yes	PD

* 'Non-CR/non-PD' is preferred over 'stable disease' for non-target disease since SD is increasingly used as an endpoint for assessment of efficacy in some trials so to assign this category when no lesions can be measured is not advised

11.4 Duration of Response

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

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

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

12 DATA REPORTING / REGULATORY REQUIREMENTS

Adverse event lists, guidelines, and instructions for AE reporting can be found in Section 7.0 (Adverse Events: List and Reporting Requirements).

12.1 Data Reporting

12.1.1 Method

This study will be monitored by the Clinical Trials Monitoring Service (CTMS). Data will be collected in the Center for Cancer Research C3D database and will be transmitted to CTMS electronically at least once every 2 weeks.

Note: **All** adverse events that have occurred on the study, including those reported through CTEP-AERS, must be reported via the monitoring method identified above.

12.1.2 Responsibility for Data Submission

N/A

12.2 CTEP Multicenter Guideline

N/A

12.3 Collaborative Agreements Language

The agent(s) supplied by CTEP, DCTD, NCI used in this protocol is/are provided to the NCI under a Collaborative Agreement (CRADA, CTA, CSA) between the Pharmaceutical Company(ies) (hereinafter referred to as "Collaborator(s)") and the NCI Division of Cancer Treatment and Diagnosis. Therefore, the following obligations/guidelines, in addition to the provisions in the "Intellectual Property Option to

Collaborator" (http://ctep.cancer.gov/industryCollaborations2/intellectual_property.htm) contained within the terms of award, apply to the use of the Agent(s) in this study:

12.3.1 Agent(s) may not be used for any purpose outside the scope of this protocol, nor can Agent(s) be transferred or licensed to any party not participating in the clinical study. Collaborator(s) data for Agent(s) are confidential and proprietary to Collaborator(s) and shall be maintained as such by the investigators. The protocol documents for studies utilizing Agents contain confidential information and should not be shared or distributed without the permission of the NCI. If a copy of this protocol is requested by a patient or patient's family member participating on the study, the individual should sign a confidentiality agreement. A suitable model agreement can be downloaded from: http://ctep.cancer.gov.

12.3.2 For a clinical protocol where there is an investigational Agent used in combination with (an)other Agent(s), each the subject of different Collaborative Agreements, the access to and use of data by each Collaborator shall be as follows (data pertaining to such combination use shall hereinafter be referred to as "Multi-Party Data"):

12.3.3 a. NCI will provide all Collaborators with prior written notice regarding the existence and nature of any agreements governing their collaboration with NCI, the design of the proposed combination protocol, and the existence of any obligations that would tend to restrict NCI's participation in the proposed combination protocol.

b. Each Collaborator shall agree to permit use of the Multi-Party Data from the clinical trial by any other Collaborator solely to the extent necessary to allow said other Collaborator to develop, obtain regulatory approval or commercialize its own Agent.

c. Any Collaborator having the right to use the Multi-Party Data from these trials must agree in writing prior to the commencement of the trials that it will use the Multi-Party Data solely for development, regulatory approval, and commercialization of its own Agent.

12.3.4 Clinical Trial Data and Results and Raw Data developed under a Collaborative Agreement will be made available to Collaborator(s), the NCI, and the FDA, as appropriate and unless additional disclosure is required by law or court order as described in the IP Option to Collaborator (http://ctep.cancer.gov/industryCollaborations2/intellectual_property.htm). Additionally, all Clinical Data and Results and Raw Data will be collected, used and disclosed consistent with all applicable federal statutes and regulations for the protection of human subjects, including, if applicable, the Standards for Privacy of Individually Identifiable Health Information set forth in 45 C.F.R. Part 164.

12.3.5 When a Collaborator wishes to initiate a data request, the request should first be sent to the NCI, who will then notify the appropriate investigators (Group Chair for Cooperative Group studies, or PI for other studies) of Collaborator's wish to contact them.

12.3.6 Any data provided to Collaborator(s) for Phase 3 studies must be in accordance with the guidelines and policies of the responsible Data Monitoring Committee (DMC), if there is a DMC for this clinical trial.

12.3.7 Any manuscripts reporting the results of this clinical trial must be provided to CTEP by the Group office for Cooperative Group studies or by the principal investigator for non-Cooperative Group studies for immediate delivery to Collaborator(s) for advisory review and comment prior to submission for publication. Collaborator(s) will have 30 days from the date of receipt for review. Collaborator shall have the right to request that publication be delayed for up to an additional 30 days in order to ensure that Collaborator's confidential and proprietary data, in addition to Collaborator(s)'s intellectual property rights, are protected. Copies of abstracts must be provided to CTEP for forwarding to Collaborator(s) for courtesy review as soon as possible and preferably at least three (3) days prior to submission, but in any case, prior to presentation at the meeting or publication in the proceedings. Press releases and other media presentations must also be forwarded to CTEP prior to release. Copies of any manuscript, abstract and/or press release/ media presentation should be sent to:

E-mail: ncicteppubs@mail.nih.gov

The Regulatory Affairs Branch will then distribute them to Collaborator(s). No publication, manuscript or other form of public disclosure shall contain any of Collaborator's confidential/ proprietary information.

12.4 Genomic Data Sharing Plan

The NIH Genomic Data Sharing (GDS) Policy does not apply to this protocol as we will be performing exploratory genetic analysis of no more than 26 patient samples; therefore, this study does not meet GDS criteria, and no GDS plan has been developed.

13 STATISTICAL CONSIDERATIONS

13.1 Study Design/Endpoints

13.1.1 Primary Endpoints

The primary objectives are:

- To establish the safety and tolerability of single agent AZD1775 in patients with refractory solid tumors.
- To determine the pharmacokinetics of AZD1775 in patients with refractory

solid tumors.

13.1.2 Secondary Endpoint

The secondary objective is:

- To evaluate the antitumor activity of AZD1775 in patients with refractory solid tumors.

13.1.3 Exploratory Endpoints

The exploratory objectives are:

- To determine the effect of AZD1775 on pharmacodynamic markers of DNA damage and apoptosis in tumor tissue and blood.
- To assess whether sufficient Wee1 inhibition is maintained throughout the therapeutic regimen.
- To identify tumor genomic alterations and gene expression patterns potentially associated with AZD1775 antitumor activity.

13.2 Study Design

This Phase I study will use a standard 3+ 3 design, and permits intra-patient dose modification (defined in Section 5.1). The MTD or recommended Phase II dose is the dose level at which no more than 1 of 6 patients experience DLT during the first cycle of treatment, and the dose below that at which at least 2 (of < 6) patients have DLT as a result of the drug.

13.3 Sample Size/Accrual Rate

With Amendment E (dated 09/24/2013), an additional expansion arm of 6 patients with documented tumors harboring BRCA- 1 or-2 mutations will also be enrolled at the MTD to further explore the safety of AZD1775 and obtain preliminary evidence of activity in this patient population. This information may inform patient selection for further development of this agent.

With Amendment G (dated 07/17/2014) an additional Arm B will be established to further evaluate an alternative once-daily dosing schedule. Patients will be enrolled based on the standard 3+3 design. Once the maximum tolerated daily dosing is established, up to 20 additional patients will be enrolled at this dose to further evaluate PD endpoints. With that number, and a tumor biopsy QA criteria failure rate of 40% with respect to paired (pre- and post-dose) biopsies, we have an 87% likelihood of having at least 10 usable PD samples, and 94% likelihood of having at least 9 usable samples. Biopsy tissue quality will be monitored and accrual will stop once we have obtained 10 usable paired samples.

The study is a standard design using 3 patients per arm, unless DLT is noted, in which

point up to 6 patients may be enrolled in an arm. Up to 30 patients will be enrolled for determination of the MTD. In addition, an additional 6 patients will be enrolled at the MTD to further define the dose and evaluate PD studies at this dose level (Expansion Arm A). To allow for a small number of patients who may not be evaluable, and the addition of Arm B, the accrual ceiling for this trial is set at 72 patients. It is anticipated that 2-3 patients per month may be enrolled onto this study. Depending on the number of patients needed to reach MTD, it is expected that 24-48 months will be required to accrue the number of patients necessary to complete the trial.

13.4 Analysis of Exploratory Endpoints

We will assess the effect of AZD1775 on DNA damage response based on immunofluorescence analysis of markers of DNA damage, Rad51, pNbs1, and γH2AX in tumor biopsies, pTyr15-Cdk, and caspase 3 in CTCs as a marker of apoptosis.

13.5 Reporting and Exclusions

13.5.1 Evaluation of toxicity – All patients will be evaluable for toxicity from the time of their first treatment with study agents.

13.5.2 Evaluation of response – All patients included in the study will be assessed for response to treatment, even if there are major protocol treatment deviations or if they are ineligible. Each patient will be assigned one of the following categories: 1) complete response, 2) partial response, 3) stable disease, 4) progressive disease, 5) early death from malignant disease, 6) early death from toxicity, 7) early death because of other cause, or 9) unknown (not assessable, insufficient data). [Note: By arbitrary convention, category 9 usually designates the "unknown" status of any type of data in a clinical database.]

All of the patients who met the eligibility criteria (with the possible exception of those who received no study medication) will be included in the main analysis of the response rate. Patients in response categories 4-9 will be considered to have a treatment failure (disease progression). Thus, an incorrect treatment schedule or drug administration does not result in exclusion from the analysis of the response rate. Precise definitions for categories 4-9 will be protocol specific.

All conclusions will be based on all eligible patients. Sub-analyses may then be performed on the basis of a subset of patients, excluding those for whom major protocol deviations have been identified (e.g., early death due to other reasons, early discontinuation of treatment, major protocol violations, etc.). However, these sub-analyses may not serve as the basis for drawing conclusions concerning treatment efficacy, and the reasons for excluding patients from the analysis will be clearly reported. The 95% confidence intervals will also be provided.

14 HUMAN SUBJECTS PROTECTIONS

14.1 Justification for Subject Selection

This study will be open to all individuals regardless of gender, ethnicity, or race, provided that the aforementioned inclusion and exclusion criteria are met. Patients for this study will be recruited through internal referral, our physician referral base, and through various cancer information hotlines (i.e., Clinical Studies Support Center, 1-800-4Cancer). To date, there is no information that suggests that differences in drug metabolism or effect on tumor would be expected in one ethnic group compared to another. Efforts will be made to extend accrual to each representative population, but a balance must be struck between participant safety considerations and limitations on the number of individuals exposed to potentially ineffective treatments on the one hand and the need to explore racial/ethnic aspects of clinical research on the other hand. If differences in outcome that correlate to ethnic identity are noted, a follow-up study may be written to investigate those differences more fully.

Due to lack of knowledge of the effects of AZD1775 on the fetus or infants, as well as the possibility of teratogenic effects, pregnant and nursing women will be excluded from this trial. Patients with unstable or serious medical conditions are excluded due to the possibility that AZD1775 may worsen their condition and the likelihood that the underlying condition may obscure the attribution of adverse events to AZD1775. HIV-positive patients on combination antiretroviral therapy are excluded from the study because of possible PK interactions with AZD1775.

14.1.1 Participation of Children

This study includes patients 18 years of age and older. Because insufficient dosing or adverse event data are currently available on the use of AZD1775 in patients <18 years of age, children are excluded from this study, but may be eligible for future pediatric trials. Studies will be performed in patients <18 years of age when it is appropriate to do so.

14.2 Evaluation of Benefits and Risks/Discomforts

There may or may not be any clinical benefit to a patient from participation in this trial. Their participation will benefit future cancer patients. Potential risks include the possible occurrence of any of a range of side effects that are listed in the consent document. The procedure for protecting against or minimizing risks will be to medically evaluate patients as described in Sections 5 and 6. Although no compensation is available, any injury will be fully evaluated and treated in keeping with the benefits or care to which participants are entitled under applicable regulations.

All patients > 18 years old will be offered the opportunity to assign a substitute decision maker on the “NIH Advance Directive for Health Care and Medical Research Participation” form, so that another person can make decisions about their medical care if they become incapacitated or cognitively impaired. The process for determining whether

adults who lose capacity to give consent may continue participation in this trial, with its heavy reliance on non-therapeutic endpoints and collection of research samples, will be on a case-by case basis and will include consultation among the substitute decision maker, protocol PI, and patient's physician regarding the health of the patient and likelihood of benefit from continued treatment.

14.3 Consent and Assent Process and Documentation

An associate or principal investigator on the trial will inform patients of the purpose, alternatives, drug administration plan, research objectives, and follow-up of this trial. The patient will be provided an IRB-approved consent for review and signature and his/her questions will be answered. After a decision is made to enroll into the study, a signature will be obtained from the patient. The original signed consent goes to Medical Records; a copy will be placed in the research record.

All patients must have a signed informed consent form and an on-study (confirmation of eligibility) form filled out and signed by a participating investigator before entering on study.

14.4 Procedure for Protecting Against or Minimizing Any Potential Risks

All care will be taken to minimize side effects, but they can be unpredictable in nature and severity. This study may involve risks to patients, which are currently unforeseeable. All patients will be monitored for side effects from taking study medication. This research represents a greater than minimal risk to participants, but presents the prospect of direct benefit to individual subjects.

The research component of this study required to obtain up to 3 CT tumor biopsies confers radiation exposure at an effective dose of 2.4 rem. This dose is below NIH RSC guidelines and represents a slightly greater than minimal risk to patients.

14.5 Patient Advocate

The patients' rights representative is available to patients receiving treatment on this protocol at the NIH Clinical Center at (301) 496-2626 in Building 10 of the Clinical Research Center, Room 1-3521, on the Bethesda NIH campus. Patients will be informed that they can contact the study PI or RN at any time with questions about their medical care, and that the patients' rights representative is also available to answer non-medical questions about the study.

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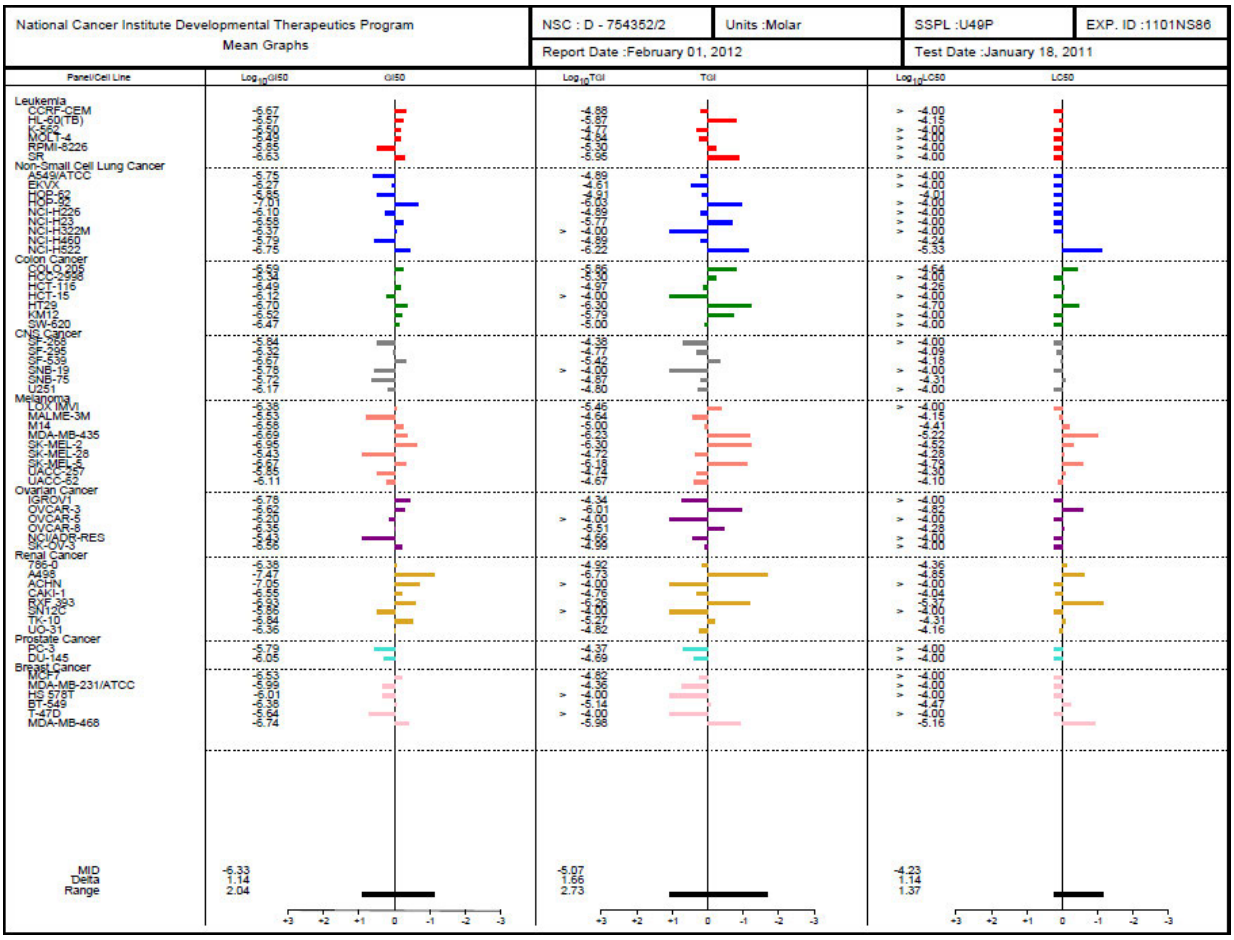
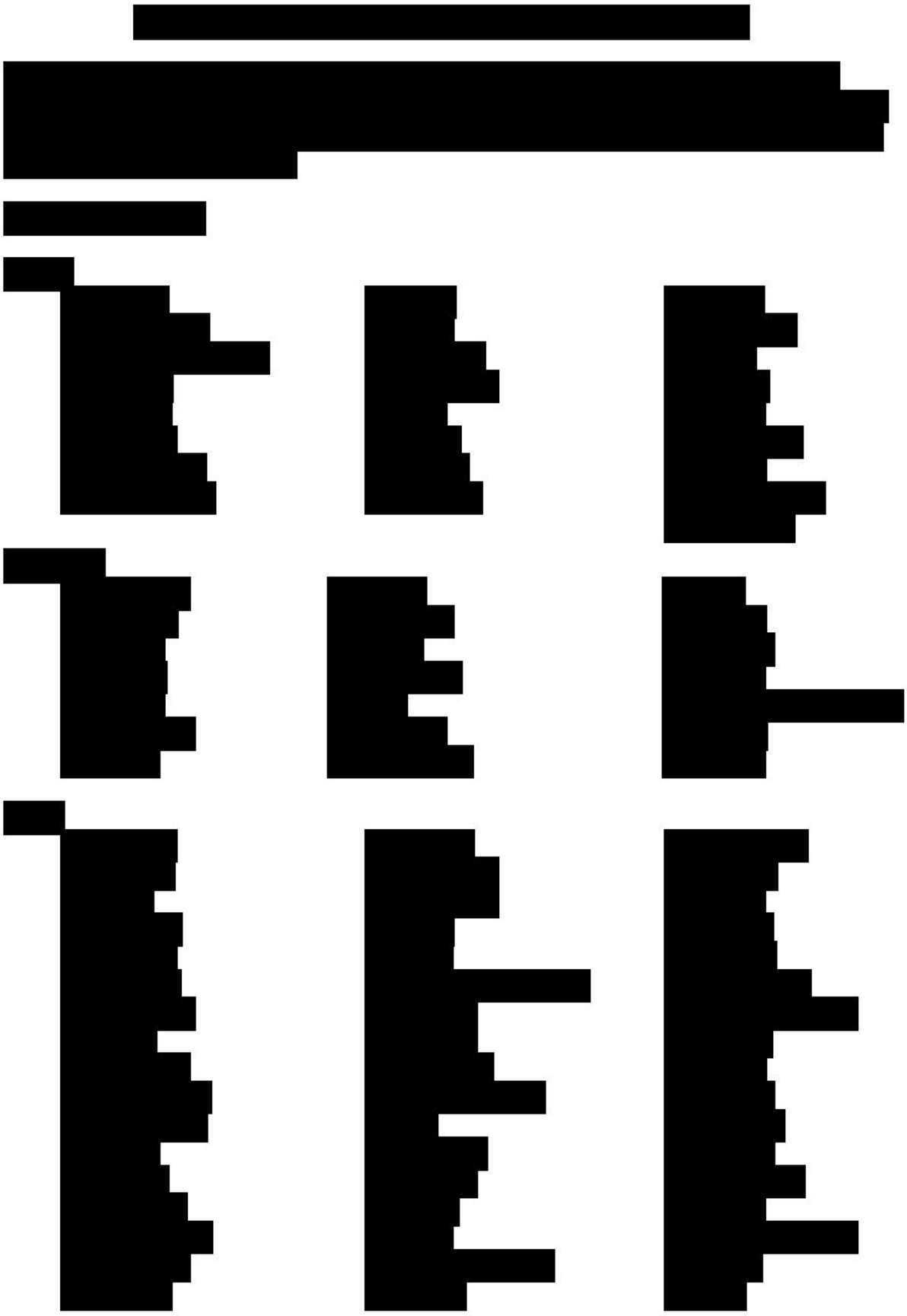
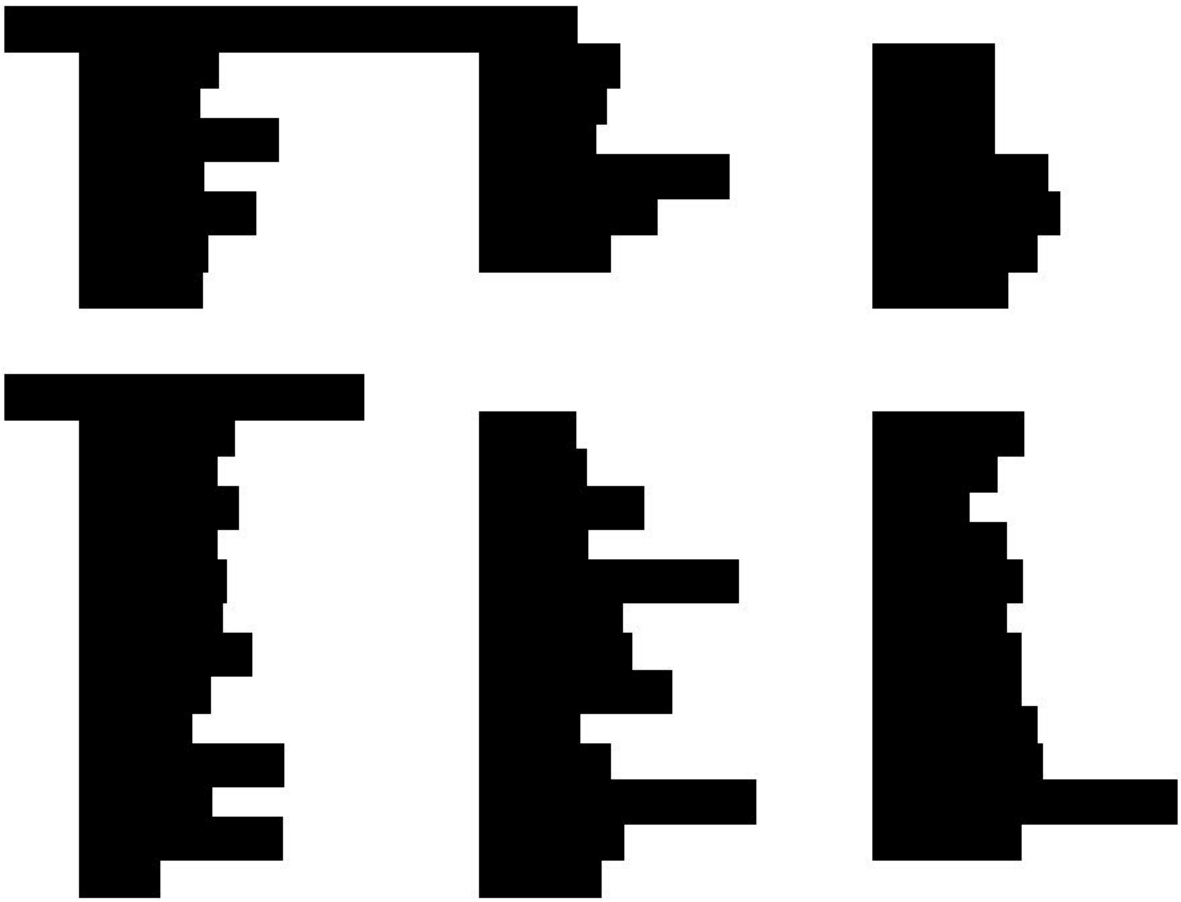
1. APPENDIX A: PERFORMANCE STATUS CRITERIA

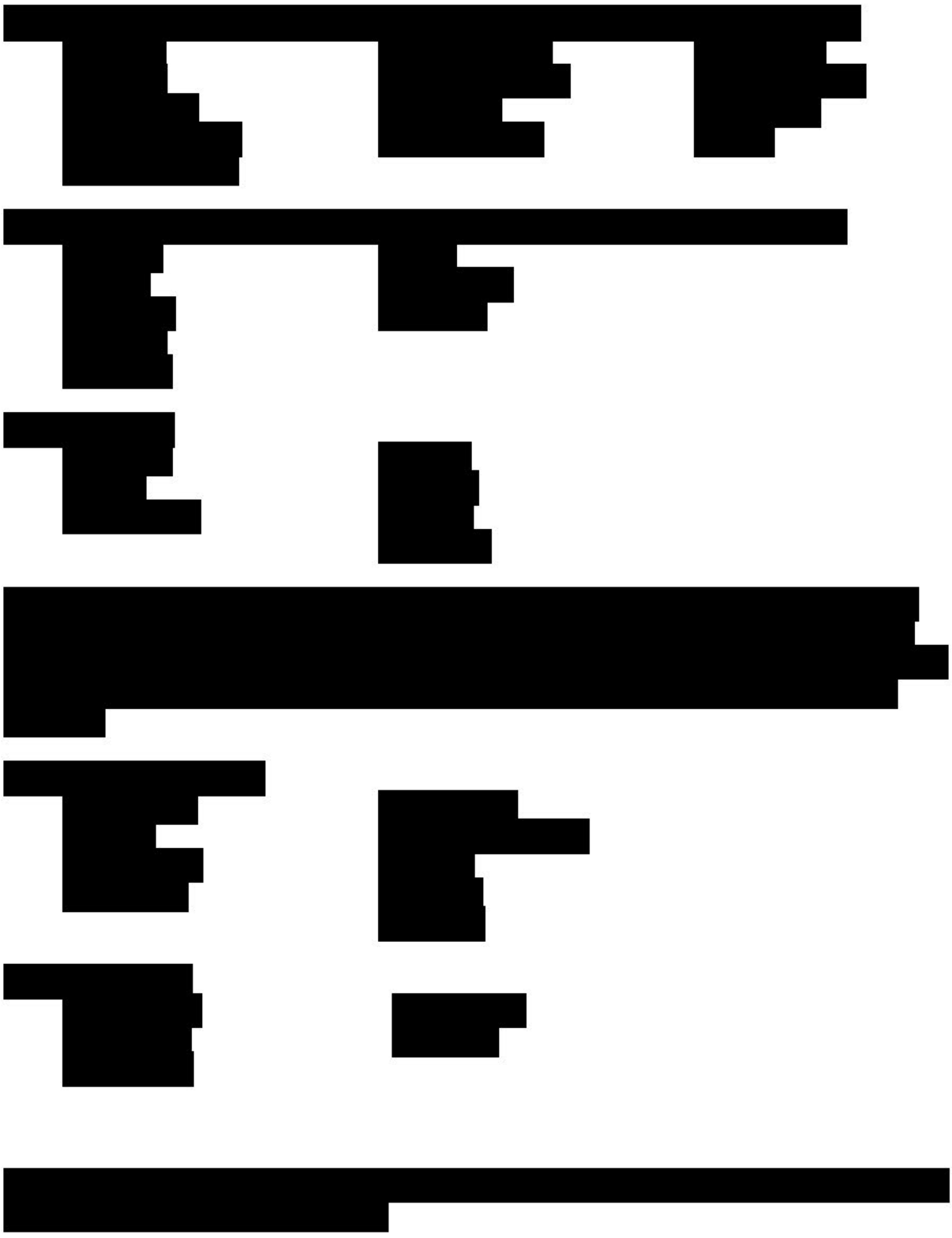
ECOG Performance Status Scale		**Karnofsky Performance Scale**	
Grade	Descriptions	Percent	Description
0	Normal activity. Fully active, able to carry on all pre-disease performance without restriction.	100	Normal, no complaints, no evidence of disease.
		90	Able to carry on normal activity; minor signs or symptoms of disease.
1	Symptoms, but ambulatory. Restricted in physically strenuous activity, but ambulatory and able to carry out work of a light or sedentary nature (*e.g.*, light housework, office work).	80	Normal activity with effort; some signs or symptoms of disease.
		70	Cares for self, unable to carry on normal activity or to do active work.
2	In bed <50% of the time. Ambulatory and capable of all self-care, but unable to carry out any work activities. Up and about more than 50% of waking hours.	60	Requires occasional assistance, but is able to care for most of his/her needs.
		50	Requires considerable assistance and frequent medical care.
3	In bed >50% of the time. Capable of only limited self-care, confined to bed or chair more than 50% of waking hours.	40	Disabled, requires special care and assistance.
		30	Severely disabled, hospitalization indicated. Death not imminent.
4	100% bedridden. Completely disabled. Cannot carry on any self-care. Totally confined to bed or chair.	20	Very sick, hospitalization indicated. Death not imminent.
		10	Moribund, fatal processes progressing rapidly.
5	Dead.	0	Dead.

2. APPENDIX B: AZD1775 (MK-1775) Activity in the NCI-60 Cell Line Screen









4. APPENDIX D: PATIENT'S MEDICATION DIARY - AZD1775

Arm A DL1 and DL-1 of AZD1775 only

INSTRUCTIONS

1. Complete one form for each cycle of treatment.

2. Swallow each capsule whole with a full glass of water either 2 hours before or 2 hours after a meal. Do not chew or open the capsules. If capsule is broken and the powder of the capsules gets on skin, wash the exposed area with as much water as necessary. Inform investigator or nurse if that occurs.

3. Record the date and time you took the capsule. The boxes in gray reflect times when you should **not** be taking drug.

4. If you have any comments or notice any side effects, please record them in the Comments column.

5. Please bring this form and your bottle of AZD1775 when you return for your Day 21 appointment.

6. In case of errors, please place a single slash mark through the error and initial it. Please do not white out any error or scribble it out with ink. Please do not write the correct information directly over the error, but on a separate line next to the error.

Today's Date _____ **Cycle #**_____ **Dose**_____**mg x5/week**

Patient Name_____ *(initials acceptable)* **Patient Study ID** _____

DL1 and DL-1 of AZD1775 only

Day	Date	Time of dose		Number of Capsules Taken			Comments
		AM	PM	25 mg	100 mg	200 mg	
1							
2							
3							
4							
5							
6							
7							
8							
9							
10							
11							
12							
13							
14							
15							
16							
17							
18							
19							
20							
21							

Patient's signature: _____

APPENDIX D - PATIENT'S MEDICATION DIARY - AZD1775

Arm A DL2 of AZD1775 and Higher

INSTRUCTIONS

1. Complete one form for each cycle of treatment.

2. Swallow each capsule whole with a full glass of water either 2 hours before or 2 hours after a meal. Do not chew or open the capsules. If capsule is broken and the powder of the capsules gets on skin, wash the exposed area with as much water as necessary. Inform investigator or nurse if that occurs.

3. Record the date and time you took the capsule. The boxes in gray reflect times when you should **not** be taking drug.

4. If you have any comments or notice any side effects, please record them in the Comments column.

5. Please bring this form and your bottle of AZD1775 when you return for your Day 21 appointment.

6. In case of errors, please place a single slash mark through the error and initial it. Please do not white out any error or scribble it out with ink. Please do not write the correct information directly over the error, but on a separate line next to the error.

Today's Date _____ **Cycle #**_____ **Dose 225 mg x 10 doses/2 weeks**

Patient Name_____ (*initials acceptable*) **Patient Study ID** _____

DL2 of AZD1775 and Higher

Day	Date	Time of dose		Number of Capsules Taken			Comments
		AM	PM	25 mg	100 mg	200 mg	
1							
2							
3							
4							
5							
6							
7							
8							
9							
10							
11							
12							
13							
14							
15							
16							
17							
18							
19							
20							
21							

Patient's signature: _____

APPENDIX D - PATIENT'S MEDICATION DIARY - AZD1775

Arm B

INSTRUCTIONS

1. Complete one form for each cycle of treatment.
2. Swallow each capsule whole with a full glass of water either 2 hours before or 2 hours after a meal. Do not chew or open the capsules. If capsule is broken and the powder of the capsules gets on skin, wash the exposed area with as much water as necessary. Inform investigator or nurse if that occurs.
3. Record the date and time you took the capsule. The boxes in gray reflect times when you should **not** be taking drug.
4. If you have any comments or notice any side effects, please record them in the Comments column.
5. Please bring this form and your bottle of AZD1775 when you return for your Day 21 appointment.
6. In case of errors, please place a single slash mark through the error and initial it. Please do not white out any error or scribble it out with ink. Please do not write the correct information directly over the error, but on a separate line next to the error.

CTEP # 9350
Clinical Center # 13-C-0032

Today's Date _____ **Cycle #**_____

Patient Name_____ (*initials acceptable*) **Patient Study ID** _____

Arm B - AZD1775

Day	Date	Time of dose	Number of Capsules Taken			Comments
			25 mg	100 mg	200 mg	
1						
2						
3						
4						
5						
6						
7						
8						
9						
10						
11						
12						
13						
14						
15						
16						
17						
18						
19						
20						
21						

Patient's signature: _____

5. APPENDIX E: INFORMATION ON POSSIBLE INTERACTIONS WITH OTHER AGENTS FOR PATIENTS AND THEIR CAREGIVERS AND NON-STUDY HEALTH CARE TEAM

Information for Patients, Their Caregivers, and Non-Study Healthcare Team on Possible Interactions with Other Drugs and Herbal Supplements

The patient _____ is enrolled on a clinical trial using the experimental study drug **AZD1775** (formerly known as MK-1775). This clinical trial is sponsored by the National Cancer Institute. This form is addressed to the patient, but includes important information for others who care for this patient.

These are the things that you as a prescriber need to know:

To the patient: Take this paper with you to your medical appointments and keep the attached information card in your wallet.

AZD1775 may interact with other drugs which can cause side effects. For this reason, it is very important to tell your study doctors of any medicines you are taking before you enroll onto this clinical trial. It is also very important to tell your doctors if you stop taking any regular medicines, or if you start taking a new medicine while you take part in this study. When you talk about your current medications with your doctors, include medicine you buy without a prescription (over-the-counter remedy), or herbal supplements such as St. John's Wort. It is helpful to bring your medication bottles or an updated medication list with you.

Many health care providers can write prescriptions. You must tell all of your health care providers (doctors, physician assistants, nurse practitioners, or pharmacists) you are taking part in a clinical trial.

These are the things that you and they need to know:

AZD1775 must be used very carefully with other medicines that need certain **liver enzymes or transport proteins to be effective or to be cleared from your system.** Before you enroll onto the clinical trial, your study doctor will work with your regular health care providers to review

any medicines and herbal supplements [redacted] Herbal medications should be avoided throughout the study.

- Please be very careful! Over-the-counter drugs (including herbal supplements) may contain ingredients that could interact with your study drug. Speak to your doctors or pharmacist to determine if there could be any side effects.
- Your regular health care provider should check a frequently updated medical reference or call your study doctor before prescribing any new medicine or discontinuing any medicine. Your study doctor's name is _____ and he or she can be contacted at _____

February 2017

STUDY DRUG INFORMATION WALLET CARD

You are enrolled on a clinical trial using the experimental study drug **AZD1775.** This clinical trial is sponsored by the NCI. **AZD1775** may interact with drugs that are **processed by your liver, or use certain transport proteins in your body**. Because of this, it is very important to:

- Tell your doctors if you stop taking any medicines or if you start taking any new medicines.
- Tell all of your health care providers (doctors, physician assistants, nurse practitioners, or pharmacists) that you are taking part in a clinical trial.
- Check with your doctor or pharmacist whenever you need to use an over-the-counter medicine or herbal supplement.

AZD1775 interacts with **CYP 3A4, 2C19, OATP1B1, OATP1B3, MATE1, MATE2K, P-gp, and BCRP** and must be used very carefully with other medicines that interact with these enzymes and proteins.

- Before you enroll onto the clinical trial, your study doctor will work with your regular health care providers to review any medicines and herbal supplement [redacted] Herbal medications should be avoided.
- Before prescribing new medicines, your regular prescribers should go to a frequently-updated medical reference for a list of drugs to avoid, or contact your study doctor.
- Your study doctor's name is _____ and he/she can be contacted at _____

6. APPENDIX F: PK/PD COLLECTION WORKSHEET

Date:	**PK/PD SAMPLE COLLECTION SHEET ARM B: Cycle 1 Day 1**				
CTEP Protocol P9350		**Ht:**	**Page for Sample Pick-up Lab phone: 301-451-1169**		**Research Nurse:** Phone: Pager:
Dose level:	**Dose AZD1775:**	**Wt:**			**PI: A. P. Chen, MD** Phone: 301-768-2749
Patient ID:		**BSA:**			
*******PK SAMPLES GO ON ICE********					
PLEASE LABEL EACH TUBE WITH ACTUAL DATE AND TIME OF SAMPLE COLLECTION					

Day	Time	Instructions	Ideal Time	Actual Time	Record comments (i.e., if collection missed), and sign each time you collect a sample
Day 1	**Prior to AZD1775 administration**	**PK 200** EDTA (lavender top) 6 mL Label tube: drug, draw date and time **PLACE ON ICE**	-----		
Day 1	**Prior to AZD1775 administration**	**PD 300** 1x 6 mL NaHep Label tube: drug, draw date and time **Room temperature**			
Day 1	**Prior to AZD1775 administration**	**PD 400** 2 x 4 mL EDTA Label tubes: drug, draw date and time **Room temperature**			
Day 1	**Prior to AZD1775 administration**	**PD 800** 1x 7 mL SST Label tube: drug, draw date and time **Room temperature**			
Administer AZD1775, Time:					
Day 1	**2 hours after dose**	**PK 201** EDTA (lavender top) 6 mL Label tube: drug, draw date and time **PLACE ON ICE**			
Day 1	**4 hours after dose**	**PK 202** EDTA (lavender top) 6 mL Label tube: drug, draw date and time **PLACE ON ICE**			

Date:	**PK/PD SAMPLE COLLECTION SHEET ARM B Cycle 1 Day 4**				
CTEP Protocol P9350 **Dose level:** **Dose AZD1775:** **Patient ID:**		**Ht:** **Wt:** **BSA:**	**Page** **for Sample Pick-up** **Lab phone: 301-451-1169**	**Research Nurse:** Phone: Pager: **PI: A. P. Chen, MD** Phone: 301 768-2749	
*******PK SAMPLES GO ON ICE********					
PLEASE LABEL EACH TUBE WITH ACTUAL DATE AND TIME OF SAMPLE COLLECTION					
Day	**Time**	**Instructions**	**Ideal Time**	**Actual Time**	**Record comments (i.e., if collection missed), and sign each time you collect a sample**
Day 4	**4 ± 1 hours after dose**	**PD 401** 2 x 4 mL EDTA Label tubes: drug, draw date and time **Room temperature**			
Administer AZD1775, Time:					

Date:	**PK/PD SAMPLE COLLECTION SHEET ARM B Cycle 1 Day 8**				
CTEP Protocol P9350 **Dose level:** **Dose AZD1775:** **Patient ID:**		**Ht:** **Wt:** **BSA:**	**Page** **for Sample Pick-up** **Lab phone: 301-451-1169**	**Research Nurse:** Phone: Pager: **PI: A. P. Chen, MD** Phone: 301 768-2749	
*******PK SAMPLES GO ON ICE********					
PLEASE LABEL EACH TUBE WITH ACTUAL DATE AND TIME OF SAMPLE COLLECTION					
Day	**Time**	**Instructions**	**Ideal Time**	**Actual Time**	**Record comments (i.e., if collection missed), and sign each time you collect a sample**
Day 8	**Prior to AZD1775 administration**	**PK 203** EDTA (lavender top) 6 mL Label tube: drug, draw date and time **PLACE ON ICE**			
Day 8	**Prior to AZD1775 administration**	**PD 402** 2 x 4 mL EDTA Label tubes: drug, draw date and time **Room temperature**			
Day 8	**Prior to AZD1775 administration**	**PD 301** 1x 6 mL NaHep Label tube: drug, draw date and time **Room temperature**			
Day 8	**Prior to AZD1775 administration**	**PD 801** 1x 7 mL SST Label tubes: drug, draw date and time **Room temperature**			
Day 8	**4 ± 1 hours after dose**	**PD 403** 2 x 4 mL EDTA Label tubes: drug, draw date and time **Room temperature**			
Administer AZD1775, Time:					

Date:	**PK/PD SAMPLE COLLECTION SHEET ARM B Cycle 1 Day 11**				
CTEP Protocol P9350 **Dose level:** **Dose AZD1775:** **Patient ID:** *******PK SAMPLES GO ON ICE********		**Ht:** **Wt:** **BSA:**	**Page for Sample Pick-up Lab phone: 301-451-1169**	**Research Nurse:** Phone: Pager: **PI: A. P. Chen, MD** Phone: 301 768-2749	
PLEASE LABEL EACH TUBE WITH ACTUAL DATE AND TIME OF SAMPLE COLLECTION					
Day	**Time**	**Instructions**	**Ideal Time**	**Actual Time**	**Record comments (i.e., if collection missed), and sign each time you collect a sample**
Day 11	**4 ± 1 hours after dose**	**PD 404** 2 x 4 mL EDTA Label tubes: drug, draw date and time **Room temperature**			
Administer AZD1775, Time:					

CTEP # 9350
Clinical Center # 13-C-0032

*****CTC samples collected at each cycle. Sample numbering to be consecutive starting from PD 405 and included below.**

Date:	**PD SAMPLE COLLECTION SHEET ARM B Cycle Day 1**				
CTEP Protocol P9350 **Dose level:** **Dose AZD1775:** **Patient ID:**		**Ht:** **Wt:** **BSA:**	**Page _____** **for Sample Pick-up** **Lab phone: 301-451-1169**		**Research Nurse:** Phone: Pager: **PI: A. P. Chen, MD** Phone: 301 768-2749
PLEASE LABEL EACH TUBE WITH ACTUAL DATE AND TIME OF SAMPLE COLLECTION					
Day	**Time**	**Instructions**	**Ideal Time**	**Actual Time**	**Record comments (i.e., if collection missed), and sign each time you collect a sample**
Day 1	**Prior to AZD1775 administration**	**PD 40** 2 x 4 mL EDTA Label tubes: drug, draw date and time **Room temperature**			
Administer AZD1775, Time:					

*****CTC samples collected at each cycle. Sample numbering to be consecutive from previous specimen and included below.**

Date:		**PD SAMPLE COLLECTION SHEET ARM B Cycle __ Day 4**			
CTEP Protocol P9350 **Dose level:** **Dose AZD1775:** **Patient ID:**		**Ht:** **Wt:** **BSA:**	**Page for Sample Pick-up Lab phone: 301-451-1169**	**Research Nurse:** Phone: Pager: **PI: A. P. Chen, MD** Phone: 301 768-2749	
PLEASE LABEL EACH TUBE WITH ACTUAL DATE AND TIME OF SAMPLE COLLECTION					
Day	**Time**	**Instructions**	**Ideal Time**	**Actual Time**	**Record comments (i.e., if collection missed), and sign each time you collect a sample**
Day 4	**4 ± 1 hours after dose**	**PD 40_** 2 x 4 mL EDTA Label tubes: drug, draw date and time **Room temperature**			
Administer AZD1775, Time:					

*****CTC samples collected at each cycle. Sample numbering to be consecutive from previous specimen and included below.**

Date:	**PD SAMPLE COLLECTION SHEET ARM B Cycle Day 8**				
CTEP Protocol P9350 **Dose level:** **Dose AZD1775:** **Patient ID:**		**Ht:** **Wt:** **BSA:**	**Page _____** **for Sample Pick-up** **Lab phone: 301-451-1169**	**Research Nurse:** Phone: Pager: **PI: A. P. Chen, MD** Phone: 301 768-2749	
PLEASE LABEL EACH TUBE WITH ACTUAL DATE AND TIME OF SAMPLE COLLECTION					
Day	**Time**	**Instructions**	**Ideal Time**	**Actual Time**	**Record comments (i.e., if collection missed), and sign each time you collect a sample**
Day 8	**Prior to AZD1775 administration**	**PD 40** 2 x 4 mL EDTA Label tubes: drug, draw date and time **Room temperature**			
Day 8	**4 ± 1 hours after dose**	**PD 40** 2 x 4 mL EDTA Label tubes: drug, draw date and time **Room temperature**			
Administer AZD1775, Time:					

*****CTC samples collected at each cycle. Sample numbering to be consecutive from previous specimen and included below.**

Date:	**PD SAMPLE COLLECTION SHEET ARM B Cycle Day 11**				
CTEP Protocol P9350 **Dose level:** **Dose AZD1775:** **Patient ID:**		**Ht:** **Wt:** **BSA:**	**Page _____** **for Sample Pick-up** **Lab phone: 301-451-1169**	**Research Nurse:** Phone: Pager: **PI: A. P. Chen, MD** Phone: 301 768-2749	
PLEASE LABEL EACH TUBE WITH ACTUAL DATE AND TIME OF SAMPLE COLLECTION					
Day	**Time**	**Instructions**	**Ideal Time**	**Actual Time**	**Record comments (i.e., if collection missed), and sign each time you collect a sample**
Day 11	**4 ± 1 hours after dose**	**PD 40** 2 x 4 mL EDTA Label tubes: drug, draw date and time **Room temperature**			
Administer AZD1775, Time:					

CTEP # 9350
Clinical Center # 13-C-0032

*****CTC samples collected at restaging. Sample numbering to be consecutive from previous specimen and included below.**

Date:	**PD SAMPLE COLLECTION SHEET ARM B RESTAGING DATE TBD**			
CTEP Protocol P9350		**Ht:**	**Page _____ for Sample Pick-up**	**Research Nurse:** Phone:
Dose level:	**Dose AZD1775:**	**Wt:**	**Lab phone: 301-451-1169**	Pager:
Patient ID:		**BSA:**		**PI: A. P. Chen, MD**
		Phone: 301 768-2749		

PLEASE LABEL EACH TUBE WITH ACTUAL DATE AND TIME OF SAMPLE COLLECTION

Day	Time	Instructions	Ideal Time	Actual Time	Record comments (i.e., if collection missed), and sign each time you collect a sample
restaging (after completion of Cycle ___)	**Prior to AZD1775 administration**	**PD 4** 2 x 4 mL EDTA Label tubes: drug, draw date and time **Room temperature**			
Administer AZD1775, Time:					