Abbreviated Title: Molecular Profiling NSCLC
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TITLE: Pilot Trial of Molecular Profiling and Targeted Therapy for Advanced Non-Small Cell Lung Cancer, Small Cell Lung Cancer, and Thymic Malignancies

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Drug Sponsor: CTEP, NCI
CTEP Protocol #: 8639

Study Drug	AZD6244	MK-2206	Lapatinib	Erlotinib	Sunitinib
NSC #	748727	749607	727989	718781	736511
IND #	77782	109493	70252	63383	Commercial Supply

NOTE: Trial administratively completed with CTEP 03/22/2016; trial remains open under NIH Intramural IRB for molecular profiling arm only

PRÉCIS

Background

- A better understanding of the genetic make-up of the individual tumor may offer potentially improved therapies. This approach may also give rapid access to response data in patients with sometimes rare genetic abnormalities.
- In addition, it will allow us to test targeted therapies in a select population of patients that is more likely to have a favorable response based on their molecular profile and the specific mechanism of action of the drug being tested.
- This approach will also speed up drug development and potentially approval, and rescue an otherwise ineffective drug candidate for the specific subgroup that can benefit.

Primary Objectives

- To determine the feasibility of the use of tumor's molecular profiling and targeted therapies in the treatment of advanced stage non-small cell lung cancer (NSCLC), small cell lung cancer (SCLC) and thymic malignancies.
- To estimate the response rate of molecular-profile directed treatments in NSCLC, SCLC and thymic malignancies patients.

Eligibility

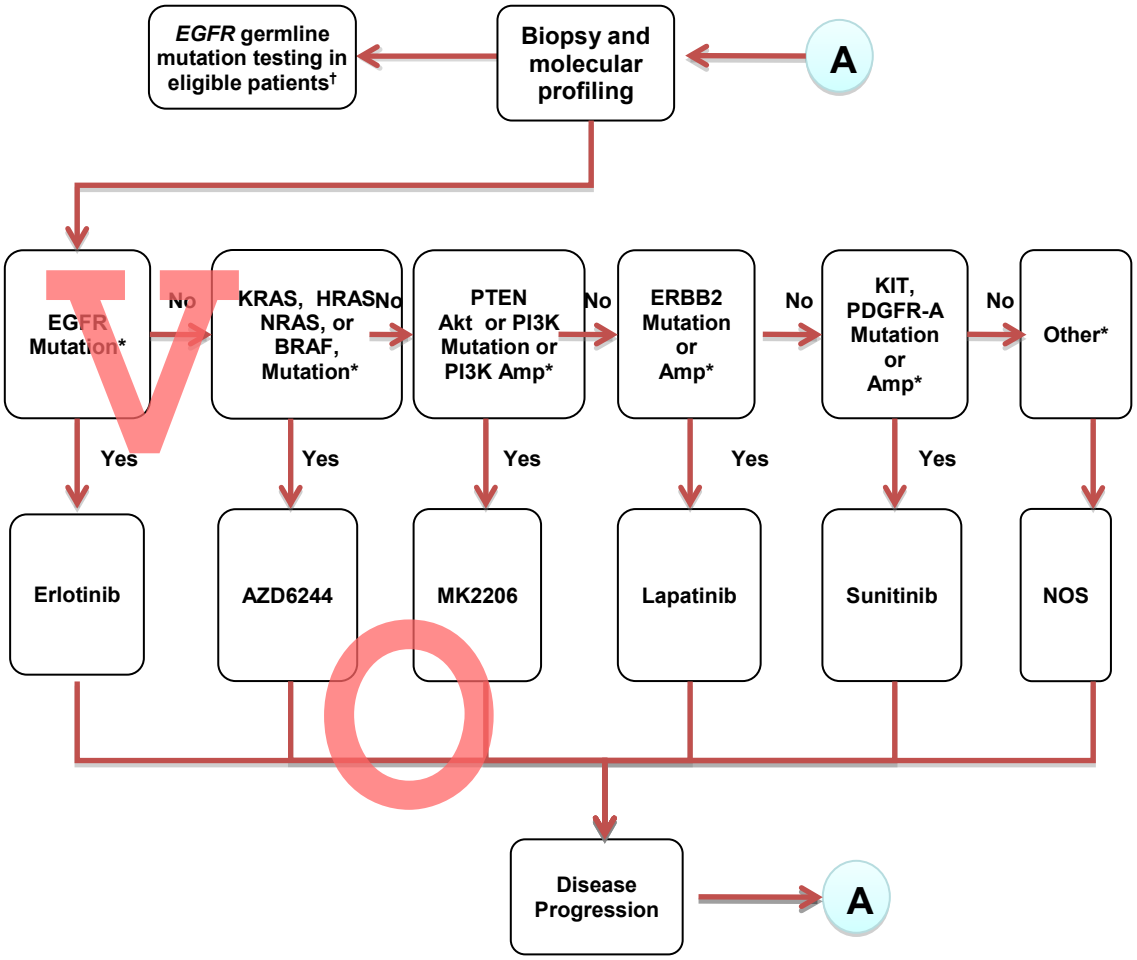
- Patients with histologically confirmed advanced lung cancer or thymic malignancies for whom surgical resection with curative intent is not feasible.
- Patients must either have biopsiable disease and be willing to undergo biopsy for molecular profiling or have paraffin embedded tissue blocks suitable for molecular profiling analysis.
- Individuals are eligible for *EGFR* germline mutation testing if they have:
    - a personal history of invasive lung cancer or one of the pre-invasive histologies associated with the development of lung cancer and more than two affected family members with invasive lung cancer or one of the pre-invasive histologies associated with the development of lung cancer; OR
    - a first-degree relative with a known *EGFR* germline mutation (*EGFR* exon 20 T790M, exon 21 V843I, exon 21 R831C and exon 20 R776G).

Design

- All patients will have their tumors undergo molecular profiling. Based on these results and on other eligibility criteria, the patients will be offered enrollment into different targeted therapy arms according to the schema shown below.
- At the NCI site only, individuals eligible for *EGFR* germline mutation will undergo testing for germline mutations affecting the *EGFR* gene; if a mutation is detected, their first-degree relatives would be invited to undergo testing for the index germline mutation found in the proband and appropriate follow-up on trial.
- Effective with Amendment I, the participating site, OHSU, will discontinue new enrollments and data entry for existing patients on the NOS arm on this protocol in favor of the OHSU protocol L8639, "Personalized Cancer Medicine Registry." The data from these patients will be included with the data from 11-C-0096 (8639) NCI patients at the

time of publication. Any OHSU patients who are eligible for a treatment arm will continue to be enrolled and followed per protocol.

Schema



*** See Sections 3.2 and 3.3 for eligibility criteria for the treatment arms. Patients will be allowed to crossover to different treatment arms as long as they meet eligibility criteria.**

† See separate eligibility criteria (Section 3.4) and schema for *EGFR* germline mutation testing.

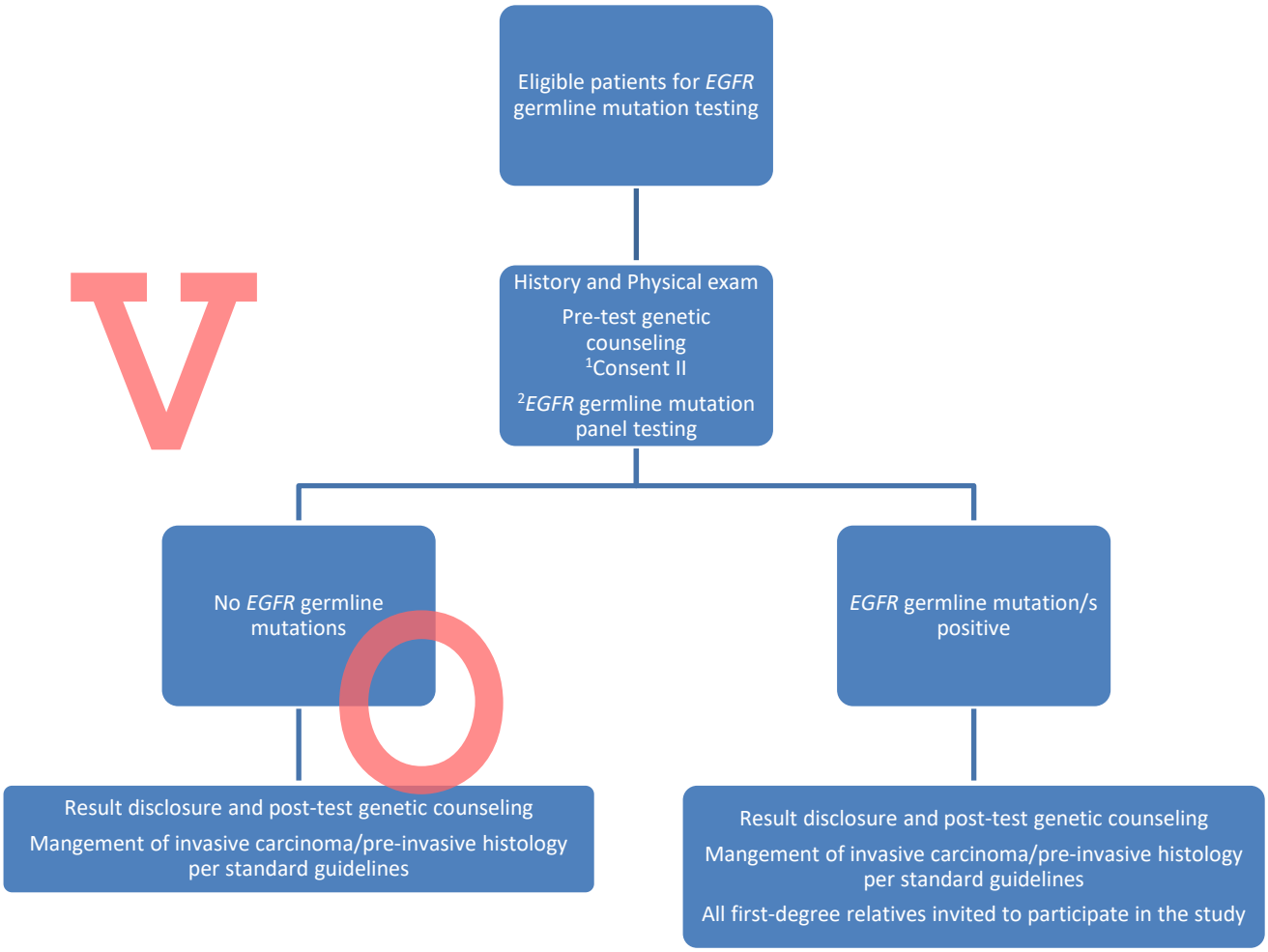
No = No or not available

Amp = gene amplification by FISH

NOS = non-otherwise specified arm

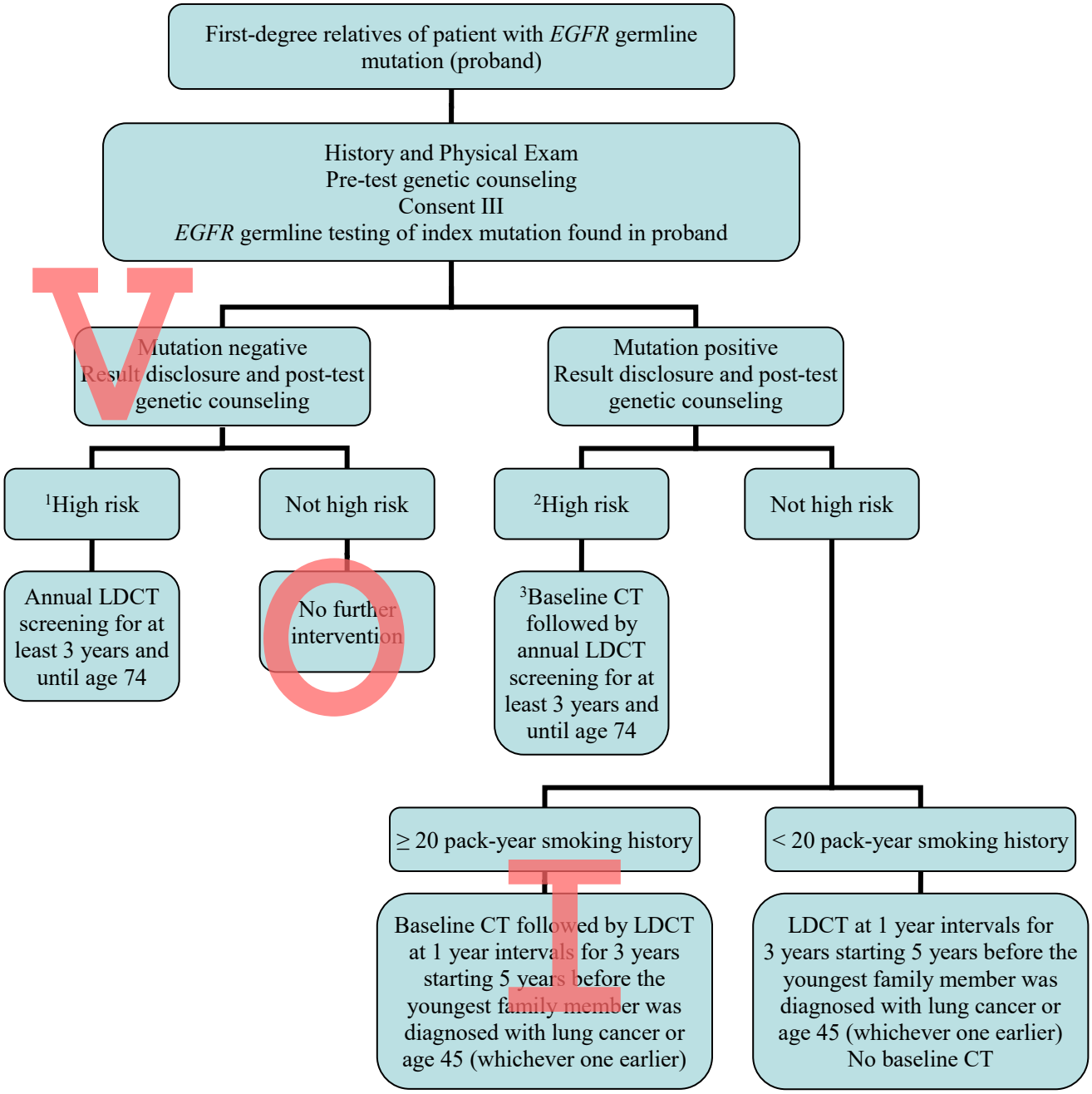
A = optional re-biopsy for patients that responded to previous line of therapy

Schema: *EGFR* Germline Mutation Testing



[1] Consent II: Informed consent for testing for *EGFR* germline mutations of proband.

[2] *EGFR* germline mutation panel: *EGFR* exon 20 T790M, exon 21 V843I, exon 21 R831C and exon 20 R776G.



[1] Between 55 and 74 years of age at enrollment with a history of cigarette smoking of at least 30 pack-years, and, if former smokers, those who quit within the previous 15 years.

[2] Age ≥ 45 years at enrollment with ≥ 20 pack-year smoking history or age within 5 years before the youngest family member was diagnosed with lung cancer.

[3] For lung nodules and incidental lesions detected on CT, follow up is per standard guidelines.

Consent III: Informed consent for testing for *EGFR* germline mutations in first-degree relatives of individuals with known *EGFR* germline mutations.

Abbreviation: LDCT, low-dose screening computed tomography scan.

Follow-up of lung nodules recommended by Fleischner Society (1) and National Comprehensive Cancer Network(2)(2)(2).

Table of Contents

Table of Tables

1 OBJECTIVES

1.1 Primary Protocol Objectives

1.1.1 To determine the feasibility of the use of tumor's molecular profiling and targeted therapies in the treatment of advanced stage non-small cell lung cancer (NSCLC), small cell lung cancer (SCLC) and thymic malignancies.

1.1.2 To estimate the response rate of molecular-profile directed treatments in NSCLC, SCLC and thymic malignancies patients.

1.2 Secondary Protocol Objectives

1.2.1 To determine the progression free survival (PFS), duration of response and overall survival (OS) of patients with NSCLC, SCLC and thymic malignancies who undergo molecular profiling and receive molecular-profile directed treatments.

1.2.2 To evaluate changes in the tumor's molecular profile on serial biopsies when patients progress after an initial response to treatment.

1.2.3 To identify molecular profiles in patients with NSCLC, SCLC and thymic malignancies and characterize their natural histories, clinical course and response to treatments.

1.2.4 To determine the frequency of *EGFR* germline mutations in families with high susceptibility to lung cancer.

2 BACKGROUND

2.1 Background and Rationale

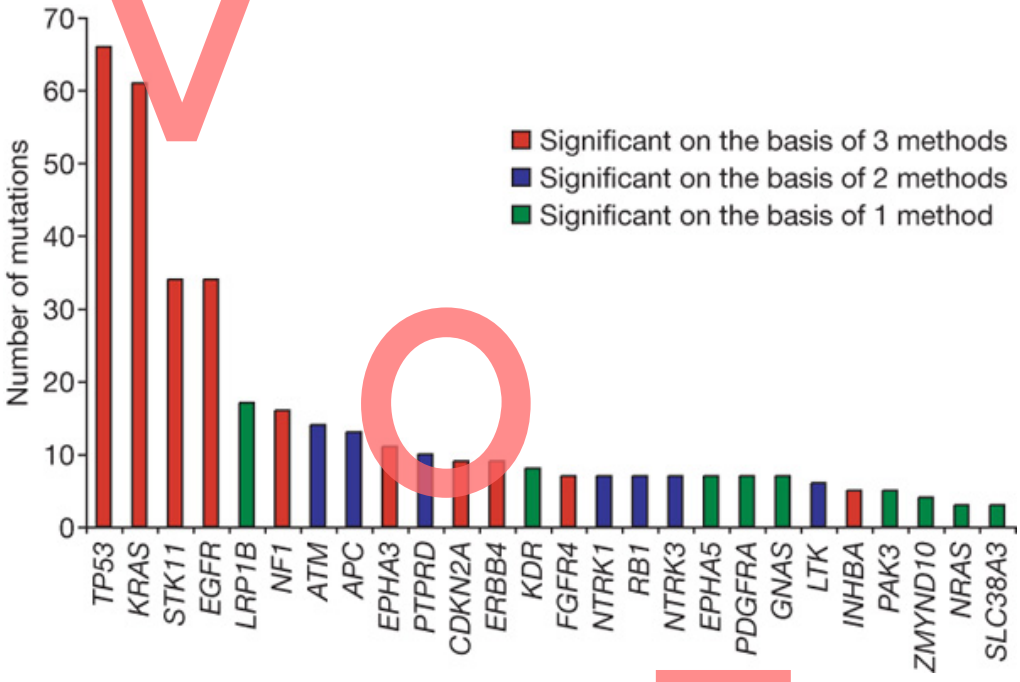
2.1.1 Background and rationale for the use of molecular profiling and molecularly based treatment in NSCLC, SCLC and thymic malignancies.

The current management of lung cancer and thymic malignancies is primarily based on the broad classification of tumors into categories based on histopathology and clinical stage at presentation. However, the existing classifications and staging systems have remained largely unchanged for decades and it seems that they have reached their limit in providing critical information that may influence management strategy. This is evident by the small improvement in overall survival of patients with this disease in the last few decades.

The heterogeneity of lung cancer and thymic malignancies at each disease stage with respect to outcome and treatment response suggests that additional subclassification and substaging remains possible. Molecular heterogeneity within individual lung cancer diagnostic categories is evident in the variable presence of specific mutations, deletions of tumor suppressor genes, and numerous chromosomal abnormalities found to date (3, 4). Reports that some of these genetic aberrations are prognostic factors for patients provide evidence that additional information on risk of relapse or death from cancer may be defined at molecular levels (4). Molecular-profiling studies indicate that activating mutations in the epidermal growth factor receptor (EGFR) and KRAS genes are generally nonoverlapping and identifiable in approximately 30 to 40% of non–small-cell lung cancers. Rarer oncogene mutations have been reported in less than 5% of lung cancer patients including mutations in ERBB2, ERBB4, MET, MEK, PI3K, AKT, FGFR1-4, and PTEN, among others (3). In addition, a fusion gene, EML4/ALK has been reported in about 6 to 13% of NSCLC patients (5). These mutations, plus others that contribute to tumor progression

("driver" mutations), can be found in almost half of non–small-cell lung cancers. Thus, correlations of molecular profiles from individual tumor samples to clinical outcome data hold the promise of a better classification of lung cancer, improved diagnostic and prognostic information and ultimately influence patient management and improve outcomes (6).

Figure 1. Significantly mutated genes in lung adenocarcinomas



Molecular characterization of thymic malignancies has also shown several abnormalities in EGFR, ERBB2, TGF and AKT among others (7-10). These abnormalities have been shown to correlate with response to targeted therapies in some cases (8, 11-13). Additionally, oncogene mutations have also been reported in small cell lung cancer patients including mutations in TP53, KDR, KIT, BCL-2, SRC, VHL, PTCH1, VHL, ERBB2, ERBB3, CDKN2A, BRAC2, ERCC1, AKT, PIK3C3, and PTEN (Table 1. Top 5 mutated genes in SCLC

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Table 1. Top 5 mutated genes in SCLC

(COSMIC database: www.sanger.ac.uk/genetics/CGP/cosmic)

Gene Name	Sample Number	Positive Samples	Percent Mutated
RB1	121	70	57%

TP53	78	62	79%
PTEN	246	34	13%
EGFR	298	16	5%
PIK3CA	99	11	11%

Evidence that molecular profiling can have a significant impact on patient's management can be found in a recent study of molecular profiling of tumors presented at the AACR 2009 conference (Von Hoff, et al. AACR 2009). This study was conducted in patients with advanced cancers that were progressing despite several previous treatments and was designed to determine whether molecular profiling of patients' tumors at this stage in their disease could provide any clinical benefit. To be eligible, patients had to undergo or have available a tumor biopsy for molecular profiling. Patients' tissue samples were submitted for molecular profiling with oligonucleotide microarray gene expression assays. The primary objective was to compare progression free survival (PFS) using a treatment regimen selected by molecular profiling with the PFS for the most recent regimen on which the patient progressed. Of 86 patients, 66 received treatments based on molecular profiling and a significant number of patients had a clinical benefit with a longer PFS treated by molecular profiling results than the PFS they had on their prior treatment regimen. This prospective study demonstrated that it is feasible to measure molecular targets in patients' tumors and that this approach may provide clinical benefit for some patients.

A better understanding of the genetic make-up of the individual tumor may offer potentially improved therapies. This approach may also give rapid access to response data in patients with sometimes rare genetic abnormalities. In addition, it will allow us to test targeted therapies in a select population of patients that is more likely to have a favorable response based on their molecular profile and the specific mechanism of action of the drug being tested. This approach will also speed up drug development and potentially approval, and rescue an otherwise ineffective drug candidate for the specific subgroup that can benefit.

2.1.2 Background and rationale for the use of erlotinib in patients with EGFR TKIs sensitizing mutations.

EGFR is a member of the receptor tyrosine kinases (RTKs) family, known as the ErbB or human epidermal receptor (HER) family of transmembrane receptors. EGFR is activated when specific ligands, such as EGF, bind to its extracellular domain. Ligand binding induces a conformational change that leads to dimerization of the receptor with either another EGFR molecule (homodimerization) or another member of the HER family (heterodimerization). Following receptor dimerization, activation of the intrinsic protein tyrosine kinase and tyrosine autophosphorylation initiates a cascade of intracellular mitogenic signaling. The EGFR can activate several different signaling pathways, including the Ras/Raf/MEK/ERK, phospholipase C, PI3K/Akt, STAT and Src/FAK pathways. These pathways form a network of signals that are important for cell growth and proliferation [7].

The development of epidermal growth factor receptor (EGFR) inhibitors has significantly influenced the field of targeted therapeutics. A high sensitivity to EGFR tyrosine kinase inhibitors (TKIs), such as gefitinib or erlotinib, has been reported in patients with EGFR-mutated lung cancers (14-16). Activating mutations in the epidermal growth factor receptor (EGFR) (Table 2. EGFR drug-sensitive and resistant mutations

) have been found in about 15% of NSCLC patients from North America and Europe, and up to 40% of Asian NSCLC patients (15-17).

Table 2. EGFR drug-sensitive and resistant mutations

	Exon	*Mutation / Deletion*
Drug sensitive	18	G719A/C
	19	delE746-A750
	21	L858R
	21	L861Q
Drug Resistant	19	D761Y
	20	T790M
	20	D770_N771insNPG

The specific EGFR tyrosine kinase inhibitors erlotinib and gefitinib have been shown to have activity in patients with activating EGFR mutations and response and survival rates are higher compared with multi-agent chemotherapy in this population. In contrast, chemotherapy is superior in patients with wild type EGFR genes (15, 17, 18). A recent phase III, randomized, controlled clinical trial in an Asian population (IPASS) has demonstrated a considerable benefit in progression free survival with the use of gefitinib as a first-line therapy for the treatment of patients with metastatic NSCLC with an EGFR TKI sensitizing mutation compared to chemotherapy (RR = 71.2% vs 47.3%; P<0.001; hazard ratio for progression, 0.48; 95% CI, 0.36 to 0.64; P<0.001) (18). Additionally, a large, non-randomized clinical trial conducted in a primarily Caucasian population from Spain has shown outstanding results with the use of EGFR TKIs in patients with NSCLC with EGFR TKI sensitizing mutations. In this trial, 217 patients with EGFR TKI sensitizing mutations that were either chemotherapy naïve or previously treated with chemotherapy received erlotinib until disease progression. PFS and OS for these patients (14 months and 27 months, respectively) was significantly higher than historical controls for unselected populations (17).

In SCLC, EGFR mutations are a rare event that represents 5% of the samples analyzed (Table 1. Top 5 mutated genes in SCLC). Interestingly, most of these mutations appear in patients with mixed histological features of SCLC and NSCLC or in patients who have never smoked. Remarkably, the EGFR mutations observed in SCLC (L858R, G719A/C and deletions in exon 19) are similar to the active mutations in NSCLC that have been reported to be predictors of a

therapeutic response to EGFR-TKIs. Furthermore, anecdotal evidence shows efficacy with the use of TKIs in these patients (19, 20).

In thymic malignancies, only one phase II study has been conducted using anti-EGFR targeted therapies. In this study, twenty-six (female = 15, male = 11) previously treated patients with metastatic thymoma (n = 19) or thymic carcinoma (n = 7), with ECOG performance status of 0 or 1, were enrolled. Patients received gefitinib orally at 250 mg daily. There were 0 CRs, 1 PR (response duration = 5 months), and 14 SDs. Six patients had SD for > 4 months. Grade 3/4 adverse events noted were: dyspnea (grade 4, n = 1; grade 3, n = 2), fatigue (grade 4, n = 1), anemia/thrombocytopenia (grade 4, n = 1), and myocardial infarction (n = 1). After completing 3 cycles, the patient with a PR developed presumed gefitinib-induced Grade 2 pneumonitis, which resolved following steroids and discontinuation of treatment. Median time to progression (TTP) was 4 months (range, 1 - 17+). None of the 5 pts (including 1 PR), analyzed by DNA sequencing, had evidence of EGFR or KRAS mutations and there was no EGFR expression analysis.

In thymic malignancies EGFR mutations are a rare event that represents < 2% of the thymic tumors analyzed (9, 10). Remarkably, the EGFR mutations (L858R and G863D in exon 21) observed in thymic tumors are similar to the active mutations in NSCLC that have been reported to be predictors of a therapeutic response to EGFR-TKIs. Even though there are no prospective trials to demonstrate the efficacy of EGFR TKIs in patients with thymic malignancies harboring EGFR mutations, anecdotal reports suggest that these patients are in fact sensitive to EGFR TKIs(9).

2.1.3 Background and rationale for the use of AZD6244 in patients with Ras/Raf/Mek/Erk pathway mutations.

The RAS/RAF/MEK/ERK pathway regulates proliferation and survival of normal cells. Hyperactivation of this pathway results in deregulated cell proliferation and malignant transformation (21). In human tumors, RAS/RAF/MEK/ERK pathway activation is a common feature which results from unregulated activation of upstream RTKs, such as EGFR, or mutations in pathway intermediates, such as KRAS, BRAF, NRAS or NF-1 (22).

The Ras superfamily of GTP binding proteins consists of more than 150 proteins, and can be grouped into at least five families (Ras, Rho, Rab, Arf, and Ran). The Ras subfamily itself includes at least 21 members (23, 24); however, there are just three closely related human Ras genes, KRAS, HRAS and NRAS, which constitute the founding members of the RAS super family. Ras signaling requires the activation of Ras by exchange of GDP for GTP, followed by interaction of Ras-GTP with Ras-effector molecules, and inactivation of Ras by GTPase-activating proteins. This signaling pathway requires the anchorage of Ras to the inner leaflet of the plasma membrane through their C-terminal S-farnesylcysteine by either a stretch of lysine residues (KRAS 4B isoform) or S-palmitoyl moieties (NRAS, NRAS and KRAS 4A isoforms)(25). The farnesyltransferase (FTase) catalyses the post-translational addition of the C-terminal cysteine residue in a process called farnesylation which is critical for the biological function of Ras (26). Ras relays its signal downstream through a high affinity complex with Raf. Activated Raf phosphorylates and activates two MAPK kinases (MEK1 and MEK2) and activated MEK then phosphorylates ERK which in turn activates multiple nuclear transcription factors (27).

Mutations in the Ras/Raf/Mek/Erk pathway are some of the most frequently mutated oncogenes in patients with NSCLC. KRAS mutations account for 20 to 38% of NSCLC cases and NRAS and BRAF for less than 3% each (3, 28-30). KRAS mutations occur more frequently in smokers and adenocarcinoma histologies. Up to 90% of the KRAS mutations are activating point mutations localized in codons 12 and 13, and less frequently in codons 61 and 63 (31).

Although mutational activation of RAS in human cancer was first demonstrated in 1982, somatic activating mutations in BRAF were only detected in 2002. Mutationally activated BRAF is detected in melanoma (70%), colorectal (15%), papillary thyroid (40%), ovarian (30%), and non-small-cell lung cancers (NSCLCs) (3%) (32, 33). Sequence analysis of the BRAF gene associated with human cancers has identified over 30 single site missense mutations, mostly within the kinase domain in exon 15. Most of the mutations of BRAF are clustered to two regions: the glycine-rich P loop of the N lobe and the activation segment and flanking regions. A Glu for Val substitution at residue 599 in the activation segment, adjacent to the conserved DFG motif, accounts for 90% of BRAF mutations in human cancers. The V599E (a.k.a. V600E) mutant of BRAF possesses the hallmarks of a conventional oncogene. The kinase activity of this mutant protein is greatly elevated, it constitutively stimulates ERK activity *in vivo* independent of RAS, and it potently transforms NIH3T3 cells. Interestingly, the conserved regulatory phosphorylation sites within the activation segment of BRAF, Thr598 and Ser601, flank Val599, leading to the suggestion that the Glu substitution at this position functions as a phospho-mimetic. Analysis of three other oncogenic mutants of BRAF showed that they stimulate kinase activity in a manner similar to V600E (32, 34, 35). In addition, non-V-600E mutations mostly localized in exon 11 have also been described in NSCLC with different frequencies but in general accounting for less than 3% of tumor or cell line samples (32, 33, 36, 37). Interestingly, both V600E and non-V600E mutations (i.e. G469A, G466V, L597V) have been shown to be sensitive to MEK inhibition in preclinical models (NSCLC cell lines)(33).

Mutations of either BRAF, NRAS or KRAS lead to constitutive activation (phosphorylation) of their downstream target, mitogen-activated protein kinase (MAPK), also known as extracellular signal-regulated protein kinase (ERK) (29, 30, 32, 34, 35). Patients with Ras/Raf/Mek/Erk pathway mutated tumors are less likely to respond to EGFR TKIs and no specific targeted therapy has thus far demonstrated activity in this patient population (14).

Table 3. Frequency of mutated KRAS in selected tumors

(3, 38) (31).

	KRAS Incidence
Lung Cancer (subtypes)	16–38%
Adenocarcinoma	22%
Broncho-alveolar Carcinoma (BAC)	31%
Large Cell	22%
Squamous Cell	1-5%

MEK or MAPK/ERK kinase is an attractive therapeutic target because its only known substrate is ERK1/2 (39). MEK inhibitors have shown good preclinical activity in different malignancies and activating mutations in the RAS (K-and N-) and BRAF genes have been reported to identify tumors that may be sensitive to MEK inhibition (32, 34, 35, 40, 41). Additionally, preclinical models have shown that synergistic cytotoxicity is achieved when combining MEK with EGFR inhibitors(42). However, CI-1040 which was the first MEK inhibitor ever tested in humans, showed poor results in clinical trials due to low systemic exposure and rapid metabolism (39). Furthermore, a second-generation more potent oral MEK inhibitor, PD0325901, was prematurely discontinued due to unexpected toxicities (Clinicaltrials.gov identifier: NCT00174369).

AZD6244 is a potent, selective, and noncompetitive ATP inhibitor of MEK1/2 that has preclinical activity against many different tumors in both cell lines and xenograft models (40). A recent phase I trial has shown promising results in patients harboring mutations in the Ras or Raf genes(43). Of the 26 patients with samples assessable for mutational status, 10 had a single mutation in KRAS (n=5), NRAS (n=4), or BRAF (n=1). The average length of time on study for patients carrying mutations (median, 3.5 months; range, 1 to 6 months) was greater than for those without a mutation (median, 2 months; range, 1 to 4 months). Several phase II clinical trials are currently underway with an emphasis on determining the efficacy of AZD6244 in tumors harboring mutations in the Ras/Raf/Mek/Erk pathway (44).

2.1.4 Background and rationale for the use of MK2206 in patients with PI3K/Akt/mTOR pathway mutations / alterations.

The PI3K/Akt/mTOR pathway is a critical regulator of cellular growth and proliferation and is essential for oncogenic transformation. It is activated through a number of tyrosine kinase growth factor receptors such as EGFR, insulin-like growth factor-1 receptor (IGF-1R), G-protein coupled receptors, and through oncogenes such as Ras (45). PI3K stimulation and mammalian target of rapamycin (mTOR) activation as well as PTEN inhibition have been demonstrated to be intimately connected to Akt activation leading to tumor mediated angiogenesis, proliferation, and cell survival. Specifically mTOR is an intracellular serine/threonine kinase that resembles that of the PI3 kinase enzymes. Once activated, mTOR can form complexes with Raptor (mTORC1) or Rictor (mTORC2) (46). The mTORC2 complex phosphorylates and activates Akt/PKB. Concurrently, the mTORC1 complex phosphorylates and activates two key regulators of translation: S6K1 (p70S6 kinase) and 4E-BP1, which both activate ribosomal formation and translation.

In a recent study, NSCLC patient samples were evaluated by FISH for PI3K amplification with > 7 copies/cell. This study revealed PI3K amplification in 42% of lung squamous cell carcinomas (47). Addtionally, P-Akt expression and loss of PTEN expression via IHC were shown to occur in NSCLC samples (48). Specifically, P-Akt expression was noted in 49% of squamous cell carcinomas and 33% of adenocarcinomas. Similarly, loss of PTEN expression was seen in 53% of SCC and 61% of adenocarcinomas. Furthermore, these findings were associated with poorer prognosis including poor differentiation, lymph node involvement, distant metastasis, late stages, and lower cumulative survival rates (48).

Mutational analyses of the PI3K/Akt/mTOR pathway in lung cancer have revealed the presence of several mutations in this pathway. Eighty-six NSCLC cell lines and 691 NSCLC patient tumor samples were evaluated for PIK3CA mutational analysis in exon 9 and 20 and gene copy

number via PCR and real-time quantitative PCR respectively. This study reported PIK3CA mutations in 4.7% NSCLC cell lines and 1.6% of the NSCLC patient samples. PIK3CA copy number gain were revealed in 9.3% of the NSCLC cell lines and 17.1% of the NSCLC patient samples, where 33.1% were SCC and 6.2% were lung adenocarcinomas. Furthermore, cell lines expressing these mutations and copy number gains correlated to increased PI3K expression and activity (49).

Table 4. PI3K mutations in NSCLC

(49)

Exon	Amino acid change	Functional domain	NSCLC cell lines (*n* = 86)	NSCLC tumors (*n* = 691)	SCLC cell lines (*n* = 43)	ExPuSC cell lines (*n* = 3)	Total no. mutations
9	S541F	Helical	0	1*	0	0	1
9	E542K	Helical	0	2*	0	0	2
9	E545K	Helical	2	5	0	0	7
9	Q546K	Helical	1	0	0	0	1
20	M1043L	Kinase	0	1	0	0	1
20	H1047R	Kinase	1	1	0	1	3
20	H1047L	Kinase	0	2	0	1	3
Total cell lines/tumors (%)			4 (4.7%)	11* (1.6%)	0 (0%)	2 (66.7%)	

*One NSCLC tumor had both S541F and E542K mutations. For histologic subtype data of cell lines, see Table 2. Mutations in the 691 NSCLC tumors occurred in the following histologic subtypes: 5 of 249 (2.0%) squamous cell tumors, 5 of 400 (1.3%) adenocarcinomas, and 1 of 42 (2.4%) other NSCLC tumors.

An activating E17K mutation in exon 4 of the AKT1 gene has been demonstrated in a small percentage of patients (<2%) with NSCLC squamous cell subtype. The activity of the endogenous kinase carrying the E17K mutation immunoprecipitated by tumour tissue was significantly higher compared with the wildtype kinase immunoprecipitated by the adjacent normal tissue. Immunostaining or immunoblot analysis on membrane-enriched extracts indicated that the enhanced membrane localization exhibited by the endogenous E17K-AKT1 may account for the observed increased activity of mutant E17K kinase in comparison with the wild-type AKT1 from adjacent normal tissue. These mutations appear to be limited to patients with SCC subtype (50-52). Mutations in the AKT2 gene have also been described. One was a missense mutation in exon 11 [1130C-A (A377V)] in a 65 year old female with lung adenocarcinoma with an EGFR L858R mutation. The other one was a deletion in intron 10 (IVS10 + 7delG) in a 66 year old male with NSCLC squamous subtype(53). Deletions and mutations have also been found on PTEN in patients with NSCLC with a frequency of approximately 9%(54, 55). Mutations present in any of the crucial domains of this gene result in reduced phosphatase activity which affects its growth suppression functions (54).

Preclinical studies have demonstrated encouraging results with inhibition of mTOR in NSCLC (56). Furthermore, in NSCLC cell lines, everolimus, an oral mTOR inhibitor, plus gefitinib have induced a significant decrease in the activation of MAPK and mTOR signaling pathways and resulted in a growth-inhibitory effect (57). In clinical trials RAD001 monotherapy was evaluated in an unselected population of 85 advanced NSCLC patients previously treated with chemotherapy alone (stratum 1) or with chemotherapy and EGFR inhibitors (stratum 2) (58). Both patient strata had to have failed two or fewer chemotherapeutic regimens including a previous platinum agent. The ORR was 7.3% in stratum 1 and 2.3% in stratum 2, where in total

there were 4 patients achieving a PR. No CRs were recorded. Additionally, the PFS was 79 days (95% CI 57-87 days) in stratum 1 and 81 days (95% CI 51-113 days) in stratum 2.

A recent study evaluating the AKT/mTOR pathway demonstrated that 50% (15 out of 30) SCLC patient tumor samples expressed mTOR via IHC analysis. Furthermore, mTOR inhibition via RAD001 was observed to inhibit SCLC tumor growth in an *in vivo* xenograft model utilizing H-69 SCLC cells. An additional study investigated the presence of PTEN mutations in SCLC in 34 SCLC cell lines and 10 SCLC tumors. This study revealed that 18% of cell lines and 10% of tumor samples had mutations in the PTEN gene including point mutations, small fragment deletions, and homozygous deletions (59). Overall, PTEN mutations have been identified in 13% of samples according to the COSMIC database (Table 1. Top 5 mutated genes in SCLC

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In addition, a target-specific mutational search revealed mutation of the PIK3CA gene in three of 13 SCLC cell lines and two of 15 primary SCLCs. In this study, these mutant PIK3CA cDNAs were introduced into cell lines establishing artificial "PIK3CA-addicted" cells and found that Triciribine, a small-molecule inhibitor of AKT signaling that is located downstream from PIK3CA, significantly inhibited the growth and colony formation activity of these cells. Using cancer cell lines, it was further shown that PIK3CA-mutated SCLC cells are more sensitive to Triciribine than PIK3CA wild-type cells. Additionally, it was found that a cisplatin-resistant subclone of PIK3CA-mutant SCLC cells was equally sensitive to Triciribine. This study for the first time uncovered PIK3CA alterations in SCLC, and suggested that anti-AKT molecular therapy could be effective for a subgroup of SCLC, which shows activation of specific genes, such as PIK3CA mutation, and that genetic stratification of SCLC according to the activation status of individual therapeutic target pathways could be clinically beneficial, especially for chemotherapy-resistant/relapsing tumors(60).

Temsirolimus is an inhibitor of mTORC1 which binds to an intracellular protein, FKBP12, and the protein-drug complex inhibits the activity of mTOR that controls cell division. The inhibition of mTOR prevents the transcription of mRNAs and translation of proteins required for cell cycle progression from G1 to S phase by mediating the activation of p70S6K and 4E-BP12. Inhibition of mTOR activity results in a G1 growth arrest in treated tumor cells. A randomized phase II study (E1500) utilizing temsirolimus in patients with extensive stage SCLC following induction chemotherapy has been conducted to study the progression-free survival (PFS) and toxicity with 25- or 250-mg doses of temsirolimus (CCI-779). Patients with either stable or responding disease to four to six cycles of cisplatin or carboplatin plus etoposide or irinotecan were randomized between 4 and 8 weeks after completion of induction therapy to receive either 25 or 250 mg of temsirolimus intravenously every week until disease progression. Eighty-seven patients entered between January 2002 and December 2003, of whom 85 were eligible: 44 received 25 mg (arm A), and 41 received 250 mg (arm 13). The overall median follow-up time for all eligible patients was 34.6 months. Median age was 59 years (range, 39-80); 42 (49.4%) were male and 43 (50.6%) female; 12.9% had brain metastases. The overall median and 1-year PFS were 2.2 months (95% confidence interval [CI]: 1.8, 2.9) and 4.7% (95% CI: 0.2%, 9.2%), respectively. The median PFS (95% CI) for arm A was 1.9 months (1.6, 2.3); for arm B, it was 2.5 months (1.9, 3.4; p = 0.24). The median overall survival from randomization was 8 months (95% CI: 6.5, 9.5). Among the 86 patients with reported toxicities, 36 (42%) had grade 3 toxicities, the most common of which were thrombocytopenia, hypophosphatemia, and fatigue, and an additional 12 (14%) had grade 4 toxicities, the most common of which was neutropenia.

No patients experienced lethal toxicities. In conclusion, temsirolimus given to responding or stable patients with extensive-stage SCLC after induction chemotherapy did not seem to result in any prolongation in PFS compared with what has been reported in the literature. However, it is important to note is that this study was conducted in a molecularly unselected population and that correlative analysis to determine alterations in the PI3K / mTOR pathway were not conducted(61).

MK-2206 is the first allosteric AKT inhibitor to enter clinical development. As an AKT inhibitor, it is equally potent towards purified recombinant human AKT1 and AKT2 isoenzymes, and 5-fold less potent against AKT3 (Investigator's Brochure, 2008). It has an advantage of having a higher intrinsic selectivity for AKT compared to the active-site AKT-specific inhibitors, i.e., ATP-competitive AKT inhibitorswhich have been clinically evaluated thus far. *In vitro* and *in vivo* anticancer efficacy of MK-2206 was demonstrated in breast cancer cell lines and in the A2780 ovarian cancer xenograft model in nude mouse and rat, either as a single agent or in combination with erlotinib, trastuzumab, and docetaxel.

Preclinical safety assessment studies in the most sensitive species, dogs, demonstrated NOAEL (no observed adverse effect level) at a 2.5 mg/kg dose (Cmax of 365 nM and AUC0-48h of 8.52 µM·h) and NOEL for cardiovascular effects at a 1 mg/kg dose (Cmax of 92 nM and AUC0-48h of 1.59 µM. Based on these data, the human equivalent doses are 34 mg (NOEL) and 80 mg (NOAEL) for a 60-kg man.

The projected human clearance for MK-2206 is 2-4 mL/min/kg with bioavailability in the range of 50%-75%. Although preclinical safety/tolerability studies identified health risks of prolonged QTc interval and hyperglycemia associated with MK-2206, no significant hyperglycemia has been seen thus far on the phase I trial; and while Grades 1-2 QTc prolongation has been noted with MK-2206 in clinical trials, it has not been thought to be clinically significant.

Merck is completing a phase 1 clinical trial with MK-2206 as a single agent in patients with solid tumors. Preliminary data from this study suggest that the dosing regimen of 60 mg orally (PO) every other day (QOD) is well-tolerated and achieves target effects of AKT inhibition. In addition, Merck has completed Phase I testing of a weekly schedule of MK-2206 and has recommended a Phase II dose of 200 mg orally (PO) weekly. Pharmacokinetic data is available for this weekly regimen. This schedule has demonstrated less toxicity than the 60 mg PO QOD schedule, appears to have at least equal antitumor activity, and is favored by Merck for subsequent trials.

2.1.5 Background and rationale for the use of lapatinib in patients with ERBB2 mutations or amplification.

HER2 (erbB-2/neu) is a member of the erbB receptor tyrosine kinase family that also includes EGFR (HER1/erbB-1), HER3 (erbB-3), and HER4 (erbB-4). Whereas these family members usually dimerize upon ligand binding, HER2, for which no ligand is reported, exists mainly in its active conformation. HER2 readily heterodimerizes with other erbB family members and is considered to be the preferred dimerization partner for EGFR, HER3, and HER4. In addition to ligand binding, receptor dimerization can be induced by a high concentration of receptors at the plasma membrane or by kinase domain mutations, resulting in receptor activation by the transphosphorylation of tyrosine residues in the C terminus of the respective molecules. The phosphorylated residues act as docking sites for an array of downstream signaling molecules activating several biochemical pathways such as the MAPK, the PI3K/Akt/mTOR, the

phospholipase C, and the Jak/Stat signaling pathways. These signal transduction cascades in concert regulate cellular processes such as proliferation, apoptosis, angiogenesis, migration, adhesion, and differentiation(62).

Mutations in the HER2 kinase domain have been reported in lung adenocarcinomas at a relatively low frequency of 2–4%. Thus far, the majority of the HER2 mutations identified in non-small cell lung cancer (NSCLC) samples are in-frame duplications or insertions in a small 8-codon region (codons 774–781 or 775–782) on exon 20. These mutations are analogous to the duplications/insertions in the 9-codon region of exon 20 in EGFR, translating to the C terminus of the αC helix in the TK (tyrosine kinase) domain. Based on this similarity, it has been postulated that mutations in HER2 cause a shift in the helical axis that narrows the ATP binding cleft, resulting in both increased TK activity and TK inhibitor sensitivity. These HER2 mutations are independent of HER2 receptor overexpression or KRAS, NRAS, BRAF, or EGFR mutations(63-65).

Studies of ERBB2 expression in NSCLC have varied in use of antibodies and immunohistochemical technique as well as the definition of positive cases. In general, positive immunostaining for ERBB2 (IHC ≥1+) in NSCLC has been reported in 2 to 60% of cases and overexpression has been associated with poor prognosis. Discrepancies among the reported frequencies might be due to different factors, including distribution of histological types, tissue processing, criteria for the scoring and interpretation of the staining results. However, strong overexpression defined by3+ IHC or gene amplification by FISH has been reported in less than 5 % of NSCLC patients (62, 66).

Molecular characterization of ERBB2 abnormalities in thymic carcinoma has shown a high rate of ERBB2 overexpression (45%) and gene amplification (50%) and a positive correlation (85%) was detected between ERBB2 protein expression and gene amplification (Kuhn E. et al. 1st International Conference on Thymic malignancies 2009).

The clinical experience with anti-HER2 directed therapies in the thoracic malignancies such as NSCLC has been disappointing most likely as a result of poor patient selection. The Cancer and Leukemia Group B (CALGB) initiated a trial in February 2000 to determine the single-agent efficacy of trastuzumab in patients with stage IIIB or IV NSCLC who had tumors that overexpressed ERBB2. Of note is that ERBB2 overexpression was defined by 2 or 3 + positivity on immunohistochemistry analysis. Among 209 screened patients, 24 patients (11%) had tumors with 2+ or 3+ expression of HER-2. However, of the 22 patients that were treated, 21 had 2+ IHC and no FISH analysis was performed. Only one patient achieved a partial response and the trial was stopped (67). A randomized phase II trial examined the effect of adding trastuzumab to a standard chemotherapeutic combination (gemcitabine–cisplatin) in patients with HER2-positive NSCLC defined by IHC ≥1. Efficacy was similar in the trastuzumab and control arms with a response rate 36% versus 41%; median time to progression 6.3 versus 7.2 months; and median progression-free survival (PFS) 6.1 versus 7 months(68).

Lapatinib (GW572016) is an oral reversible, dual tyrosine kinase inhibitor of EGFR (ERBB1) and HER2/neu (ERBB2). A study conducted in chemotherapy naïve patients with NSCLC was stopped for futility after 131 patients were randomized. Median age 66 (range 32–86); female 56%; BAC 20%, No BAC 71%; previously untreated 98.5%; current/former smokers 70%, never smoker 30%. There were no complete responses. Of 56 patients in the target population, 1 (2%) achieved partial response (PR), 11 (20%) had stable disease (SD) of 24 wks; in the non-target

population, 1 patient had a PR (1.3%) and 12 (16%) had SD of 24 wks. Three patients had ERBB1 mutations (G719S, S768I, KRAS G12S; L858R and T790M; L858R) but none of them responded. Three of 77 patients evaluated had ERBB1 gene copy increase (none of whom responded) and 2 had ERBB2 gene copy increase (one had a 51% decrease in tumor size). The most common adverse events were grade 1/2 diarrhea, nausea, rash, vomiting and fatigue, and were similar in both groups(69).

Despite the lack of efficacy of lapatinib in an unselected population of patients with NSCLC, there is no prospective clinical data on the use of Lapatinib in patients with ERBB2 mutations or amplification. As stated above, these mutations are analogous to the duplications/insertions in the 9-codon region of exon 20 in EGFR, translating to the C terminus of the αC helix in the TK domain. Based on this similarity, it has been postulated that mutations in ERBB2 cause a shift in the helical axis that narrows the ATP binding cleft, resulting in both increased TK activity and TK inhibitor sensitivity(63-65, 70). Furthermore, preclinical studies assessing the *in vivo* effect of a dual EGFR / ERBB2 TKI (BIBW2992) on HER2(YVMA) transgenic mice or H1781 xenografts with documented tumor burden revealed that BIBW2992 has significant activity in regards to signaling and tumor shrinkage(63).

Theoretically it might be predicted that the protein expression would be superior for assessing response to trastuzumab therapy because the antibody binds to the cell surface protein. In breast cancer, however, FISH technique has been demonstrated to be a more accurate and reliable method for selecting patients eligible for treatment with anti-HER2 directed therapy(66). In NSCLC HER2 status assessed by IHC has been has been shown to be a method that is not optimal. On the other hand, anecdotal evidence supports the notion that FISH analysis would be a better method to predict the response to anti-HER2 therapies in NSCLC patients (66, 70). Moreover, in a phase II clinical trial of trastuzumab in combination with chemotherapy in NSCLC, the few patients whose tumors had HER2 gene amplification and who were treated with trastuzumab had a better response to trastuzumab compared to other patients. Response rate (83%) and median PFS (8.5 months) are higher in trastuzumab-treated patients with HER2 3+ or fluorescence in situ hybridization (FISH)-positive NSCLC compared to patients without HER2 overexpression (68).

2.1.6 Background and rationale for the use of sunitinib in patients with PDGFR-A gene amplification or mutations or KIT mutations.

The PDGFR/PDGF system includes two receptors (PDGFRA and PDGFRB) and four ligands (PDGFA, PDGFB, PDGFC, and PDGFD). Ligand binding induces receptor dimerization, enabling autophosphorylation of specific tyrosine residues and subsequent recruitment of a variety of signal transduction molecules. PDGFR regulates normal cellular growth and differentiation, and expression of activated PDGFR promotes oncogenic transformation Increased expression of PDGFR has also been associated with poor prognosis in NSCLC (71).

Analysis of early stage (I and II) NSCLC samples by IHC revealed a high expression of PDGFR-A in 51% of squamous cell carcinoma, 77% in large cell carcinoma, 73% in adenocarcinomas (73%), and 68% in bronchioalveolar carcinoma (68%) (72). However, a similar study performed in early stage NSCLC patient samples identified strong PDGFR-A staining by IHC in just 2 to 3% of cases (73). In addition, mutations in the PDGFR-A gene have been identified in 3.7% of cases of adenocarcinoma (3).

Sunitinib malate is an oral, selective multitargeted tyrosine kinase inhibitor with antiangiogenic and antitumor activities. It inhibits VEGF receptor (VEGFR)-1, -2, and -3 and PDGFR-A and -B activity, as well as the activity of several related tyrosine kinases. It is FDA approved for its use in previously untreated patients with advanced renal cell carcinoma and in imatinib-refractory gastrointestinal stromal tumors (GIST). A recent phase II clinical study has revealed modest efficacy of single-agent sunitinib in advanced NSCLC patients with an ORR of 11.1% (95% CI, 4.6% to 21.6%). An additional 18 patients (28.6%) experienced stable disease of at least 8 weeks in duration. Median progression-free survival was 12.0 weeks (95% CI, 10.0 to 16.1 weeks), median overall survival was 23.4 weeks (95% CI, 17.0 to 28.3 weeks), and therapy was generally well tolerated (74).

However, sunitinib has not been tested in selected populations of NSCLC patients who have mutations or overexpression of PDGFR or VEGFR and there is no retrospective analysis correlating the levels of these receptors with clinical outcomes(74). Using a high-throughput cancer cell line screening platform, it has been found that a small fraction of human tumor-derived cell lines show significant sensitivity to single-agent sunitinib exposure. These two cell lines [a non–small-cell lung cancer (NSCLC) and a rhabdomyosarcoma] showed expression of highly phosphorylated PDGFRA. In the sunitinib-sensitive adenosquamous NSCLC cell line, PDGFRA expression was associated with focal PFGRA gene amplification, which was similarly detected in a small fraction of squamous cell NSCLC primary tumor specimens. Moreover, in this NSCLC cell line, focal amplification of the gene encoding the PDGFR ligand PDGFC was also detected, and silencing PDGFRA or PDGFC expression by RNA interference inhibited proliferation. These findings suggest that, rare tumors that show PDGFRA activation may be more likely to be responsive to pharmacologic inhibition by sunitinib (75).

The c-kit proto-oncogene encodes a Kit transmembrane tyrosine kinase receptor whose ligand is stem cell factor (SCF), a growth factor important in the stimulation and formation of various cell types. Upon SCF binding, the Kit receptor dimerizes and autophosphorylates to in turn activate many downstream signal transduction components including PI3K, PKB/Akt, Src, Janus transcription pathway, and the Ras-Raf-MAPK cascade that all in some form promote cell survival and increase cellular proliferation. Mutations in KIT lead to a constitutive activation of the receptor in a ligand independent fashion. It has been shown through structural analyses of the KIT receptor and related tyrosine kinase receptors that the juxtamembrane domain of this receptor plays an inhibitory role on the regulation of this receptor. Furthermore, this ability to suppress Kit receptor firing is compromised by mutations and deletions seen in the juxtamembrane domain.

The presence of KIT mutations has been found in less than 4% of samples from thymic malignancies. In contrast to other mutations, KIT mutations have been found exclusively in thymic carcinomas. One of these mutations is a V560 deletion in exon 11. This mutation is associated with sensitivity to KIT inhibitors based upon multiple lines of evidence: (a) the growth of mutant-bearing Ba/F3 cells *in vitro* is readily inhibited by treatment with imatinib and sunitinib(7); (b) a patient whose tumor harbored this mutation responded to treatment with imatinib(8); and (c) this mutation has also been found in an imatinib-sensitive gastrointestinal stromal tumor (76). Another KIT-mutant case of thymic carcinoma reported in the literature harbored an L576P mutation in exon 11 (10). This mutation has also previously been described in gastrointestinal stromal tumor and melanoma and has been biologically characterized as being sensitive to imatinib (76). The third type of mutation reported is a D820E mutation in exon 17

exhibited by a thymic carcinoma responding to sorafenib(13). Finally, the KIT H697Y mutation identified in exon 14 is associated *in vitro* with sensitivity to sunitinib and imatinib (7).

2.1.7 Rationale for the non-otherwise (NOS) specified arm

In order to accomplish the primary objective of identifying molecular profiles in patients with NSCLC, SCLC, and thymic malignancies and characterize their natural histories, clinical course and response to treatment, we will follow all patients that undergo molecular profiling from the time that they are enrolled in the trial until the time of death. For this reason, we have created a study arm called the "non otherwise specified" (NOS) arm. In this arm, we will include all patients that undergo molecular profiling under this protocol, but that are otherwise not eligible for any of the active treatment arms in the protocol. This will allow us to link their clinical and mutational analyses, to determine the frequency of each mutation, its association with clinical features, response to treatment and outcome, and its association with other mutations. As future therapeutic protocols specific for these mutations are developed, patients may be notified of their eligibility for these studies, if they consent. Future translational studies may be used to: a) unravel the complex biology of lung cancer; b) identify prognostic markers; c) define predictive markers of response/resistance to new therapies; and d) identify new targets.

2.1.8 Background and rationale for *EGFR* germline mutation testing in lung cancer families

Lung cancer is the leading cause of cancer-related death worldwide, accounting for more than one million deaths every year (2, 77). Cigarette smoke accounts for approximately 85-90% of all lung cancers. Other etiological factors for lung cancer include radon (a radioactive gas produced by decay of radium 226), asbestos, lung inflammation and scarring (78).

Although the etiological role of genetic factors in lung cancer is poorly understood, several lines of evidence suggest a role for these factors in lung carcinogenesis even after adjustment for age, gender and smoking habits. For example, in the Environment And Genetics in Lung cancer Etiology (EAGLE) study, the relative risk of lung cancer associated with a positive family history, adjusted for age, gender, residence, education, and smoking, was 1.57 (95% CI 1.25-1.98) (79). Genome-wide association (GWA) studies have suggested lung cancer susceptibility loci at various locations (80). Other studies have identified single nucleotide polymorphisms (SNPs) in enzymes involved in DNA repair and detoxification of xenobiotics and environmental chemicals as well as germline mutations in pro-survival pathways (81-84). However, as cigarette smoking is such an overwhelming and preventable risk factor, the importance of family history and genetic susceptibility to lung cancer risk has often been overlooked and is difficult to discern.

Germline testing for inherited predisposition is a well established part of care of individuals who may be at hereditary risk for cancers of the breast, ovary, colon, stomach, uterus, thyroid, and other primary sites(85, 86). In contrast to somatic genetic profiling of tumor tissue, germline testing involves analysis of DNA from blood or saliva for inherited mutations in specific genes that are associated with the type of cancer seen in the individual or family seeking assessment (86).

High-penetrance germline mutations usually result in a significant alteration in the function of the corresponding gene product and are associated with large increases in cancer risk. For example, *BRCA1* mutation, which is found at a frequency of 1 in 166 to 1000 in the general population, confers a relative risk for breast cancer of 32 in women aged between 40 and 49.

Other mutations are associated with less dramatic increases in risk. *APC**I1307K and *rs10505477* at 8q24 are associated with relative risks of 1.7 (intermediate-penetrance mutations) and 1.2 respectively for colon cancer (low-penetrance variants) (86). Germline testing of several high-penetrance germline mutations are now part of standard clinical practice and identification of these mutations often justifies an adjustment of clinical care through the modification of surveillance or through preventive surgery. Genetic tests for intermediate-penetrance mutations and low-penetrance variants are of uncertain clinical utility because the cancer risk associated with the mutation is generally too small to form an appropriate basis for clinical decision making.

Table 5. Previously reported germline mutations in lung cancer, clinico-pathologic features of the patients and their response to TKI

Germline mutation	Author, year	Proband	Ethnicity	Smoking status	Stage at presentation	Histology	Concurrent somatic mutations	Response to EGFR TKI	Family History	Germline mutation in family members	Follow up
EGFR exon 20 T790M	Carmelo et al, 2011	72F	European	NS	IV	Adenocarcinoma	*EGFR* E746-A750 in exon 19	PR/ PFS 9 months	Sister with LC (below)	Absent in 2 daughters	OS 23 months
		74F	European	NS	IIIBwet	Poorly differentiated carcinoma	Negative	PR/ PFS 45 months		Absent in 2 sons	NR
	Girard et al, 2010	66F	Asian Indian	NS	IV	Mixed adenocarcinoma (acinar and bronchioloalveolar)	somatic *EGFR* L858R mutation	NR	2 family members with lung cancer in 2 generations	NR	NR
		58M	Eastern European	NS	IV	Poorly differentiated adenocarcinoma (acinar and solid)	*EGFR* L858R	NR	7 family members with lung cancer in 5 generations	NR	NR
	Bell et al, 2005	50M	European	Smoker	IV	BAC	*EGFR* L858R, del L747-T751	PFS 9 months	6 family members with lung cancer, BAC or pulmonary nodules in 3 generations	*EGFR*T790M in all 4 siblings	NR
		55M	European	NR	IV	Adenocarcinoma	*EGFR* G719A	No response			OS 6 months
	Prudkin et al, 2009	72F	NR	NS	NR	Adenocarcinoma, BAC, LCNEC	None	NR	2 family members with lung cancer in 2 generations	NR	NR
EGFR exon 21 V843I	Ikeda et al, 2008	70F	NR	NR	IIA, IA	Adenocarcinoma, BAC, AAH	*EGFR* L861Q *EGFR* L858R	Not used	3 family members with lung cancer in 2 generations	*EGFR*L861Q in 3 siblings	PFS 8 months*
	Ohtsuka et al 2010	48F	Japanese	NR	IV	Adenocarcinoma	NR	No response	4 individuals with lung cancer in 3 generations	*EGFR*V843I in 4 family members- 3 with lung adenocarcinoma and 1 healthy person	OS NR
EGFR exon 21 R831C	Chung et al, 2010	76F	NR	NS	IIIA	Adenocarcinoma	*EGFR* L861R	PR PFS 14 months	Sister died of lung cancer at 67 years	NR	
EGFR exon 20 R776G	Centeno et al, 2011	47M	European	Smoker	IIIA	Adenocarcinoma	*EGFR* exon 21 L858R	NR	One sibling died of cancer	NR	OS 1 year

Abbreviations: EGFR: epidermal growth factor; PFS: progression free survival; NR: not reported; NS: never-smoker; BAC: bronchioalceolar carcinoma; AAH: atypical adenomatous hyperplasia; LCNEC: large cell neuroendocrine carcinoma; TKI: tyrosine kinase inhibitor OS: overall survival

No high penetrance mutations have been described for lung cancer. Germline mutations in *EGFR* and its association with familial lung cancer have been rarely described, but their penetrance and patterns of familial involvement are not known. Previously described germline mutations and clinico-pathologic features of lung cancer patients are shown in Table 5. *EGFR*T790M is the most extensively studied germline mutation in familial lung cancer. The T790M mutation occurs within exon 20, which encodes part of the kinase domain and results in an amino acid substitution at position 790 in EGFR, from a threonine (T) to a methionine (M). It is known to occur as a 'second-site mutation' in more than 50% of patients who develop acquired resistance (progressive disease while on therapy after an initial response) to EGFR tyrosine kinase inhibitor (TKI) therapy (87, 88).

Bell *et al* reported a family with multiple cases of lung cancer associated with germline transmission of the *EGFR*T790M mutation (81). Four of the six tumors analyzed showed a secondary somatic activating *EGFR* mutation (either L858R, del L747-T751, or G719A) occurring in *cis* with the germ line T790M mutation. Although somatic *EGFR*T790M mutations are common in patients with acquired resistance to EGFR inhibitors, germline *EGFR*T790M mutations are rare. Two subsequent studies examined this germline mutation in a larger series of patients: Girard *et al* found *EGFR*T790M germline mutation in two of 369 cases of never smokers with NSCLC- both patients had a family history significant for lung cancer (89). However Vikis *et al* analyzed genomic DNA from 237 probands representing lung cancer families with more than three affected individuals for *EGFR*T790M mutation (90). No mutation was observed in any of the family probands analyzed and in any of 60 random fresh-frozen resected lung tumors suggesting that it may be a minor contributor to genetic susceptibility in familial lung cancers. Furthermore, in the Genetic Epidemiology of Lung Cancer Consortium (GELCC) linkage study, the analysis of 52 families (and family subsets within) did not reveal a significant logarithm of odds (LOD) score on or near 7p11, where the *EGFR* gene is located, which also suggests that the mutation may not be a major contributor to familial predisposition to lung cancer. Prudkin *et al* identified one germ line *EGFR*T790M mutation in a cohort of 240 patients with previously untreated lung adenocarcinoma (91).

EGFR exon 21V843I is an additional germline mutation which has been described in two reports, in association with a significant family history of lung cancer (92, 93). One subsequent study found no V843I mutations in 285 NSCLC patients with family history of lung cancer (94).

The role if any of these mutations in inherited susceptibility to lung cancer in families with clustering of germline mutation and lung cancer is not completely clear. However early evidence suggests that *EGFR*T790M may provide a proliferative advantage with respect to wild-type (WT) *EGFR*, possibly due to the enhanced kinase activity of the mutant (90, 95-97). In a human bronchial epithelial cell line, overexpression of *EGFR*T790M displayed a growth advantage over WT *EGFR* (90). Moreover, presence of a concurrent L858R mutation, i.e. T790M/L858R double mutation resulted in a substantial increase in phosphorylation, compared with the L858R alone (95). Additionally, even in the absence of additional kinase domain mutations, *EGFR* T790M mice developed tumors, albeit with longer latency than *EGFR* L858R/T790M mice. Enhanced tyrosine autophosphorylation was also observed in cells transfected with the plasmid containing *EGFR*R776G (a germline *EGFR* mutation identified in Case 3 of Table 5) when compared to those transfected with WT *EGFR* (94).

At this time, the clinical utility of identification of these germline mutations in lung cancer is not known. It is likely that these and other intrinsic genetic factors controlling susceptibility to lung

cancer might be hidden by strong environmental factors like cigarette smoking and air pollution which have been implicated in the mutagenesis of many other genes controlling pathways involved in the development lung cancers (3, 98). However, identification of these germline mutations may benefit individuals by providing deeper self-knowledge and motivation to pursue healthy behaviors (e.g., quit smoking), even if the results may not inform clinical decision making (99).

With Amendment F, we will begin testing selected individuals (see Section 3.4) for the presence of *EGFR* germline mutations known to be associated with susceptibility to lung cancer. Only patients who enroll at the NCI will be eligible for this testing.

2.2 CTEP-Supplied Investigational Agent(s)

See Section 18.1.1

2.3 Other agents

See Section 18.1.2

2.4 Hypothesis

A better understanding of the genetic make-up of the individual tumor may offer potentially improved therapies. This approach may also give rapid access to response data in patients with sometimes rare genetic abnormalities. In addition, it will allow us to test targeted therapies in a select population of patients that is more likely to have a favorable response based on their molecular profile and the specific mechanism of action of the drug being tested. This approach will also speed up drug development and potentially approval, and rescue an otherwise ineffective drug candidate for the specific subgroup that can benefit.

3 PATIENT SELECTION

3.1 Eligibility criteria for initial enrollment

3.1.1 Patients with histologically confirmed advanced NSCLC, SCLC and thymic malignancies for whom surgical resection or multimodality therapy with curative intent is not feasible. For patients with Stage III NSCLC, who can be encompassed by a radiation port, definitive XRT should have been performed first when possible.

3.1.2 Individuals who meet the eligibility criteria for *EGFR* germline mutation testing (Section 3.4) but who do not have advanced cancer as defined in 3.1.1 may enroll for *EGFR* germline mutation testing only and will not be eligible for the treatment or NOS arms.

3.1.3 Patients with advanced cancer must meet one of the following criteria (does not apply to first-degree relatives or individuals with pre-invasive histology enrolling only for *EGFR* germline mutation testing):

- Patients must have biopsiable disease and be willing to undergo biopsy for molecular profiling

  or

- Patients must have enough and adequate archival material from a previous biopsy to perform molecular profiling analyses. The adequacy of the material provided

will be determined by the principal investigator in conjunction with the laboratories performing the molecular profiling analyses.

or

- Patients must have previously undergone a successful molecular profiling of their tumor with mutation analysis of any of the genes described in section 5.2 or ALK break apart fluorescence in situ hybridization, as part of this protocol (crossover patients) or other molecular profiling protocols such as the Lung Cancer Mutation Consortium protocol among others.

3.1.4 Age ≥18 years.

3.2 Eligibility criteria for enrollment into the Treatment arms (Treatment arms are no longer recruiting as of 3/22/16)

Please refer to section 3.3 for eligibility criteria for enrollment in the specific molecular profile / treatment arms of this protocol.

3.2.1 Inclusion Criteria

3.2.1.1 Patients must have measurable disease, defined as at least one lesion that can be accurately measured in at least one dimension (longest diameter to be recorded) as >20 mm with conventional techniques or as >10 mm with spiral CT scan. See Section 13 for the evaluation of measurable disease.

Target lesions cannot be selected within previously irradiated areas, if not newly arising or clearly progressing after irradiation as proven by repeat scanning.

3.2.1.2 Life expectancy of greater than 3 months.

3.2.1.3 Performance status (ECOG) ≤ 2 (See **Appendix C: Performance Status Criteria**).

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

- leukocytes >1,500/mcL
- absolute neutrophil count >1,000/mcL
- platelets >100,000/mcL
- total bilirubin* < 1.5 X institutional upper limit of normal

  For patients with Gilbert's syndrome or levels of indirect bilirubin above the upper limit of normal, direct bilirubin will be used for eligibility and should be < 1.5 X institutional upper limit of normal.
- AST(SGOT)/ALT(SGPT) < 3 X institutional upper limit of normal
- creatinine ≤1.5 X institutional upper limits of normal

  OR creatinine clearance >50 mL/min/1.73 m2 for patients with creatinine levels above 1.5 X institutional normal.

3.2.1.5 Patients must have recovered from toxicity related to prior therapy (chemotherapy, surgery or radiation) to grade ≤ 1 (defined by CTCAE: The NCI Common Terminology Criteria for Adverse Events Version 4 (CTCAE) will be used for toxicity and adverse event reporting. All appropriate treatment areas have access to a copy of the CTCAE version 4.0. A copy of the CTCAE version 4.0 can be downloaded from the CTEP web site (http://ctep.cancer.gov).

3.2.1.6 The effects of most of the therapeutic agents used in this trial on the developing human fetus at the recommended therapeutic doses are unknown. For this reason and because some agents are known to be teratogenic, women of child-bearing potential and men must agree to use adequate contraception (hormonal or barrier method of birth control; abstinence) prior to study entry and for the duration of study participation, and continue for at least 16 weeks after completing the study to avoid pregnancy and/or potential adverse effects on the developing embryo. Should a woman become pregnant or suspect she is pregnant while participating in this study, she should inform her treating physician immediately. Because there is an unknown but potential risk for adverse events in nursing infants secondary to treatment of the mother with targeted therapies, breastfeeding should be discontinued if the mother is treated with targeted therapies.

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

3.2.2 Exclusion Criteria

3.2.2.1 Patients who have had major surgery, chemotherapy or radiotherapy within 2 weeks prior to entering the study or those who have not recovered from adverse events due to agents administered more than 2 weeks earlier.

3.2.2.2 Patients may not be receiving any other investigational agents or other medications for the treatment of their malignancy.

3.2.2.3 Patients with symptomatic brain metastases should be excluded from this clinical trial because of their poor prognosis and because they often develop progressive neurologic dysfunction that would confound the evaluation of neurologic and other adverse events. However, patients who have had treatment for their brain metastases and whose brain metastatic disease status has remained stable for at least 1 week after the end of brain radiation may be enrolled to undergo molecular profiling at the discretion of the principal investigator. In addition, brain metastatic disease should be stable for at least 4 weeks, before the patients can be enrolled in any of the experimental treatment arms.

3.2.2.4 Patients with any condition (e.g., gastrointestinal tract disease resulting in an inability to take oral medication or a requirement for IV alimentation, prior surgical procedures affecting absorption, or active peptic ulcer disease) that impairs their ability to swallow and retain tablets are excluded.

3.2.2.5 Any uncontrolled medical illness that precludes the patient from undergoing a biopsy for molecular profiling and / or receiving treatment under one of the experimental arms of the study should be excluded. These conditions include but are not limited to:

- Ongoing or uncontrolled, symptomatic congestive heart failure (Class III or IV as defined by the NYHA functional classification system (see **Appendix D**).
- Uncontrolled hypertension

- Unstable angina pectoris
- Cardiac arrhythmia
- Uncontrolled diabetes
- Uncontrolled psychiatric illness/social situations that would limit compliance with study requirements.

3.2.2.6 Patients with QTc prolongation (defined as a QTc interval equal to or greater than 500 msec) or other significant ECG abnormalities are excluded.

3.2.2.7 Caution should be used if patients are required to use a concomitant medication that can prolong the QT interval and efforts should be made to switch to a different medication before the patient begins treatment under an experimental arm. See **Appendix E** for a table of medications with the potential to prolong the QTc interval. A comprehensive list of agents with the potential to cause QTc prolongation can be found at: http://www.azcert.org/medical-pros/drug-lists/bycategory.cfm

3.2.2.8 The eligibility of patients taking medications that are potent inducers or inhibitors of that enzyme will be determined following a review of their case by the Principal Investigator. (A list of potent CYP3A4 inducers or inhibitors can be found in Appendix F). Every effort should be made to switch patients taking such agents or substances to other medications before they begin treatment with one of the experimental drug included in this protocol, particularly patients with gliomas or brain metastases who are taking enzyme-inducing anticonvulsant agents. A comprehensive list of medications and substances known or with the potential to alter the pharmacokinetics of sunitinib through CYP3A4 is provided in **Appendix F**.

3.2.2.9 Patients with tumor amenable to potentially curative therapy as assessed by the investigator.

3.2.2.10 Pregnant women are excluded from this study because many of the FDA approved agents and investigational agents in this trial have the potential for teratogenic or abortifacient effects. Because there is an unknown but potential risk for adverse events in nursing infants secondary to treatment of the mother with these agents, breastfeeding should be discontinued if the mother is treated in this protocol. These potential risks may also apply to other agents used in this study.

3.3 Specific eligibility criteria for enrollment in treatment arms (Treatment arms are no longer recruiting as of 3/22/16)

The following is a description of the eligibility criteria required to be enrolled in the specific molecular profile-based treatment arms of this study which include: Erlotinib, AZD6244, MK-2206, Lapatinib and Sunitinib arms. This is in addition to the general eligibility and exclusion criteria described in sections **3.1** and **3.2**.

3.3.1 Erlotinib arm

3.3.1.1 Inclusion criteria

3.3.1.1.1 Patients must have an EGFR TKI sensitizing mutation as determined by analysis of the primary tumor or a metastatic site in a CLIA certified laboratory.

3.3.1.1.2 Patients with SCLC or thymic malignancies must have been treated with at least 1 previous standard of care chemotherapy regimen or refuse to be treated with conventional chemotherapy agents.

3.3.1.2 Exclusion criteria

3.3.1.2.1 Previous anti-EGFR TKI therapy

3.3.1.2.2 Patients with a known EGFR TKI resistant mutation.

3.3.1.2.3 History of allergic reactions attributed to compounds of similar chemical or biologic composition to Erlotinib.

3.3.2 AZD6244 arm

3.3.2.1 Inclusion criteria

3.3.2.1.1 Patients must have one of the following as determined by analysis of the primary tumor or a metastatic site in a CLIA certified laboratory:

- KRAS mutation or
- NRAS mutation or
- HRAS mutation or
- BRAF mutation

3.3.2.1.2 Patients must have been treated with at least 1 previous standard of care chemotherapy regimen or refuse to be treated with conventional chemotherapy agents.

3.3.2.2 Exclusion criteria

3.3.2.2.1 Any prior exposure to MEK, Ras, or Raf inhibitors.

3.3.2.2.2 History of allergic reactions attributed to compounds of similar chemical or biologic composition to AZD6244.

3.3.2.2.3 Cardiac conditions as follows:

- Uncontrolled hypertension (BP ≥150/95 despite optimal therapy)
- Heart failure NYHA Class II or above (**See Appendix D: New York Heart Association Classifications**)
- Prior or current cardiomyopathy
- Baseline LVEF ≤ 50%.
- Atrial fibrillation with heart rate >100 bpm
- Unstable ischaemic heart disease (MI within 6 months prior to starting treatment, or angina requiring use of nitrates more than once weekly).

3.3.2.2.4 Laboratory values as listed below (SI units):

- Absolute Neutrophil Count (ANC) <1.5x109/L (1500 per mm3)
- Platelets <100x109/L (100,000 per mm3)
- Hemoglobin (Hgb) <9.0 g/dL

- Serum bilirubin >1.5 x upper limit of normal (ULN)
- Aspartate aminotransferase (AST/SGOT) or alanine aminotransferase (ALT/SGPT) >2.5 x ULN (≥5 ULN in presence of liver metastases).

3.3.3 MK2206 arm

3.3.3.1 Inclusion criteria

3.3.3.1.1 Patients must have one of the following as determined by analysis of the primary tumor or a metastatic site in a CLIA certified laboratory:

- PI3KCA mutation or
- PI3KCA gene amplification by FISH (gene to chromosome ration >2) or
- AKT mutation or
- PTEN mutation

3.3.3.1.2 Patients must have been treated with at least 1 previous standard of care chemotherapy regimen or refuse to be treated with conventional chemotherapy agents.

3.3.3.2 Exclusion criteria

3.3.3.2.1 Previous AKT inhibitor therapy.

3.3.3.2.2 History of allergic reactions attributed to compounds of similar chemical or biologic composition to MK-2206.

3.2.3.2.3 Preclinical studies demonstrated the potential of MK-2206 for induction of hyperglycemia in all preclinical species tested. Patients with diabetes or in risk for hyperglycemia should not be excluded from trials with MK-2206, but the hyperglycemia should be well controlled on oral agents before the patient enters the trial.

3.3.4 Lapatinib arm

3.3.4.1 Inclusion criteria

3.3.4.1.1 Patients must have one of the following as determined by analysis of the primary tumor or a metastatic site in a CLIA certified laboratory:

- ERBB2 mutation or
- ERBB2 gene amplification by FISH (gene to chromosome ration >2)

3.3.4.1.2 Patients must have been treated with at least 1 previous standard of care chemotherapy regimen or refuse to be treated with conventional chemotherapy agents.

3.3.4.2 Exclusion criteria

3.3.4.2.1 Previous Lapatinib therapy.

3.3.4.2.2 History of allergic reactions attributed to compounds of similar chemical or biologic composition to lapatinib.

3.3.4.2.3 LVEF < 50%

3.3.5 Sunitinib arm

3.3.5.1 Inclusion criteria

3.3.5.1.1 Patients must have one of the following as determined by analysis of the primary tumor or a metastatic site in a CLIA certified laboratory:

- PDGFR-A mutation or
- PDGFR-A gene amplification by FISH (gene to chromosome ratio >2) or
- KIT mutation

3.3.5.1.2 Patients must have been treated with at least 1 previous standard of care chemotherapy regimen or refuse to be treated with conventional chemotherapy agents.

3.3.5.2 Exclusion criteria

3.3.5.2.1 Previous sunitinib therapy

3.3.5.2.2 History of allergic reactions attributed to compounds of similar chemical or biologic composition to sunitinib.

3.3.5.2.3 Patients who require use of therapeutic doses of coumarin-derivative anticoagulants such as warfarin are excluded. Note: Low molecular weight heparin is permitted provided the patient's PT INR is <1.5.

3.3.5.2.4 Patients with any of the following conditions are excluded:

- Serious or non-healing wound, ulcer, or bone fracture.
- History of abdominal fistula, gastrointestinal perforation, or intra-abdominal abscess within 28 days of treatment.
- Any history of cerebrovascular accident (CVA) or transient ischemic attack within 12 months prior to study entry.
- History of myocardial infarction, cardiac arrhythmia, stable/unstable angina, symptomatic congestive heart failure, or coronary/peripheral artery bypass graft or stenting within 12 months prior to study entry.
- History of pulmonary embolism within the past 12 months.

3.3.5.2.5 Patients with a pre-existing thyroid abnormality who are unable to maintain thyroid function in the normal range with medication are ineligible.

3.4 Eligibility criteria for *EGFR* germline mutation testing (NCI site only)

3.4.1 Age ≥ 18 years with one of the following criteria:

3.4.2 Personal history of invasive lung cancer or one of the pre-invasive histologies associated with the development of lung cancer [adenocarcinoma in situ (AIS), minimally invasive

adenocarcinomas (MIA) or atypical adenomatous hyperplasia (AAH)] and more than two affected family members with invasive lung cancer or one of the pre-invasive histologies associated with the development of lung cancer; OR

3.4.3 First-degree relatives of an individual enrolled in the study with a known *EGFR* germline mutation; OR

3.4.4 Detection of de-novo (prior to any treatment with TKI) T790M mutation in the tumor specimen of a lung cancer patient; OR

3.4.5 A family history of lung cancer not possibly related to smoking history, defined as at least one family member within the 3rd degree of consanguinity with lung cancer who was a never or oligo-smoker* or at least two family members in either the maternal or paternal side with a history of lung cancer; OR

3.4.6 Documented germline lung cancer associated mutation (such as EGFR T790M) tested elsewhere.

*An oligo-smoker is defined as an individual with ≤ 15 pack-year history of smoking.

3.5 Inclusion of Women and Minorities

Both men and women of all races and ethnic groups are eligible for this trial. The following are our accrual targets based on race and gender for allhistology groups combined (i.e. NSCLC, SCLC, and thymic malignancies):

Accrual Targets

Ethnic Category	**Sex/Gender**				
	Females		***Males***		***Total***
Hispanic or Latino	42	+	42	=	84
Not Hispanic or Latino	258	+	258	=	516
Ethnic Category: Total of all subjects	300	+	300	=	600
Racial Category					
American Indian or Alaskan Native	12	+	12	=	24
Asian	90	+	90	=	180
Black or African American	42	+	42	=	84
Native Hawaiian or other Pacific Islander	12	+	12	=	24
White	144	+	144	=	288
Racial Category: Total of all subjects	300	+	300	=	600

Accrual Rate: 10 patients/month

Total Expected Accrual: 600 patients from all participating sites and in all diseases combined.

4 PATIENT REGISTRATION

4.1 Registration Process

4.1.1 NCI Patient Registration

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 treatment-eligible patient on the protocol prior to the release of any investigational agents. Verification of Registration will be forwarded electronically via e-mail to the research team. Please note, 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.

Patients will retain their number throughout their time on the protocol but will be given a letter designation to identify which arm they have been enrolled to, or re-enrolled to in accordance with Section **5.4**. The letters that correspond to the treatment arms are as follows:

A – Arm A (Erlotinib)

B – Arm B (AZD6244)

C – Arm C (MK-2206)

D – Arm D (Lapatinib)

E – Arm E (Sunitinib)

F – Arm F (NOS)

Version M 04/08/14 NOTE: The treatment arms are closed to accrual, but patients are still being accrued for germline testing.

4.1.2 For Participating Site Registration

NOTE: The participating site closed on 02/05/2015. This section is no longer applicable, but is being retained for historical purposes.

All patients must be registered through the NCI Central Registration Office (CRO). The CRO is open from 8:30am to 5:30pm EST Monday through Friday, excluding federal holidays. The protocol registration form and cover memo will be supplied by the Coordinating Center, NCI CCR, and updates will be provided as needed. Subject eligibility and demographic information is required for registration. To initially register a subject after the participant has signed consent, complete the top portion of the form and fax to the CRO at 301-480-0757. Once eligibility is confirmed, complete the remainder of the form which is the eligibility checklist and, fax the completed registration checklist and cover memo to the CRO at 301-480-0757.

The CRO will notify the registering site personnel either by e-mail or fax that the protocol registration form has been received. The CRO will assign a unique patient/subject ID number for each subject that will be used to enter data into the C3D database. Questions about eligibility should be directed to the Coordinating Center's Research Nurse:

Arlene Berman, RN

9000 Rockville Pike
Bldg 10, Rm 12N226
Bethesda, MD 20892
Telephone: 301.435.5609
Fax: 301.480.2590
Email: arleneb@mail.nih.gov

Technical questions about the form should be directed to the Central Registration: Office (301-402-1732).

4.2 On-Study Research Evaluation

This section applies only to patients enrolled into any of the treatment arms of the protocol.

4.2.1 Complete history and physical examination (including height, weight, vital signs and ECOG performance score) with documentation of 1) measurable disease, 2) narcotic use and pain assessment, and 3) prior therapies (surgical, radio therapeutic and cytotoxic) will be conducted prior to on-study. A complete medication history will be obtained prior to starting, including medications at base line, over the counter medications, homeopathic remedies, vitamins, and alternative therapies.

4.2.2 Imaging Studies (Baseline) - Every patient should have a baseline clinical evaluation with CT scan of chest, abdomen and/or pelvis for areas of known or suspected disease involvement prior to receiving treatment. In some patients an MRI may be more appropriate. FDG-PET scans are recommended but not mandatory. Baseline scans must be completed within 28 days prior to enrollment.

4.2.3 Laboratory Evaluation [baseline labs must be obtained within one week prior to enrollment].

4.2.4 Hematological Profile: CBC with differential and platelet count, prothrombin time, activated partial thromboplastin time.

4.2.5 Biochemical Profile: Sodium, potassium, chloride, CO2, BUN, creatinine, glucose, AST, ALT, alkaline phosphatase, bilirubin, albumin, total protein, LDH, calcium, phosphorous, magnesium, and urinalysis.

4.2.6 PT and PTT

4.2.7 EKG (baseline)

4.2.8 Pregnancy test for female patients of childbearing age and anatomic ability.

5 STUDY DESIGN / TREATMENT PLAN

The purpose of this trial is to perform molecular profiling analysis in patients with advanced NSCLC, SCLC and thymic malignancies and offer enrollment into targeted therapy arms based on their molecular profiles. We will also be studying the frequency of *EGFR* germline mutations by enrolling patients from families with high susceptibility to lung cancer and their first-degree relatives (at the NCI only).

5.1 Specimen Collection

Individuals who meet the eligibility criteria for *EGFR* germline mutation testing but who do not have advanced cancer as defined in Section 3.1.1 will not undergo tumor biopsy for molecular profiling.

Patients with advanced cancer (as defined in Section 3.1.1) who meet eligibility criteria will undergo biopsy of the primary tumor or any metastatic site for molecular profiling analyses (with the exception of patients who have available archival material or patients that have previously undergone successful molecular profiling). All attempts will be made to obtain fresh tissue biopsies but if this is not feasible, archived paraffin blocks will be acceptable as long as there is enough and adequate material to perform the molecular profiling analyses described below. The adequacy of the archival tissue provided by the patient will be determined by the principal investigator at each participating institution in conjunction with the molecular pathology laboratories performing the molecular analyses. The following techniques will be used to obtain tissue samples for molecular profiling according to the location and size of the lesions:

- Core needle biopsy
- Excisional biopsies

Sample collection will be performed either by the interventional radiology or the surgery departments at the participating institution. Biopsies will be performed according to the standard operating procedures in place at the performing institution. Samples will be formalin fixed and sent to the local pathology department for further processing and confirmation of the histologic diagnosis. If possible, one of the samples will be immediately frozen to perform some of the correlative analyses described in Section 10.

5.2 Molecular Profiling

After the tumor samples are obtained, molecular profiling analyses will be performed to determine enrollment in specific treatment arms (with the exception of patients who have previously undergone successful molecular profiling). Since these analyses will be used for treatment decisions, they will be performed in a CLIA certified laboratory at the participating institutions. The following is a list of the molecular profiling analyses to be performed:

5.2.1 Gene mutation analyses for treatment decisions

Approved Gene Symbol	Approved Gene Name	Location	Sequence Accession IDs	Previous Symbols	Aliases
AKT1	v-akt murine thymoma viral oncogene homolog 1	14q32.32-q32.33	M63167 NM_005163		RAC, PKB, PRKBA, AKT
AKT2	v-akt murine thymoma viral oncogene homolog 2	19q13.1-q13.2	NM_001626		
BRAF	v-raf murine sarcoma viral oncogene homolog B1	7q34	M95712 NM_004333		BRAF1
EGFR	epidermal growth factor receptor (erythroblastic leukemia viral (v-erb-b) oncogene homolog, avian)	7p12	NM_005228	ERBB	ERBB1

ERBB2	v-erb-b2 erythroblastic leukemia viral oncogene homolog 2, neuro/glioblastoma derived oncogene homolog (avian)	17q11.2-q12	X03363	NGL	NEU, HER-2, CD340, HER2
HRAS	v-Ha-ras Harvey rat sarcoma viral oncogene homolog	11p15.5	AJ437024 NM_176795	HRAS1	
KIT	v-kit Hardy-Zuckerman 4 feline sarcoma viral oncogene homolog	4q11-q12	S67773	PBT	CD117, SCFR, C-Kit
KRAS	v-Ki-ras2 Kirsten rat sarcoma viral oncogene homolog	12p12.1	BC010502 NM_033360	KRAS2	KRAS1
NRAS	neuroblastoma RAS viral (v-ras) oncogene homolog	1p13.2	BC005219 NM_002524		
PDGFRA	platelet-derived growth factor receptor, alpha polypeptide	4q12	D50001 NM_006206		CD140a, PDGFR2
PIK3CA	phosphoinositide-3-kinase, catalytic, alpha polypeptide	3q26.3	NM_006218		p110-alpha, MGC142161, PI3K, MGC142163
PTEN	phosphatase and tensin homolog	10q23	U92436 NM_000314	BZS, MHAM	MMAC1, TEP1, PTEN1

5.2.2 ERBB2 amplification by FISH

5.2.3 PDGFRA amplification by FISH

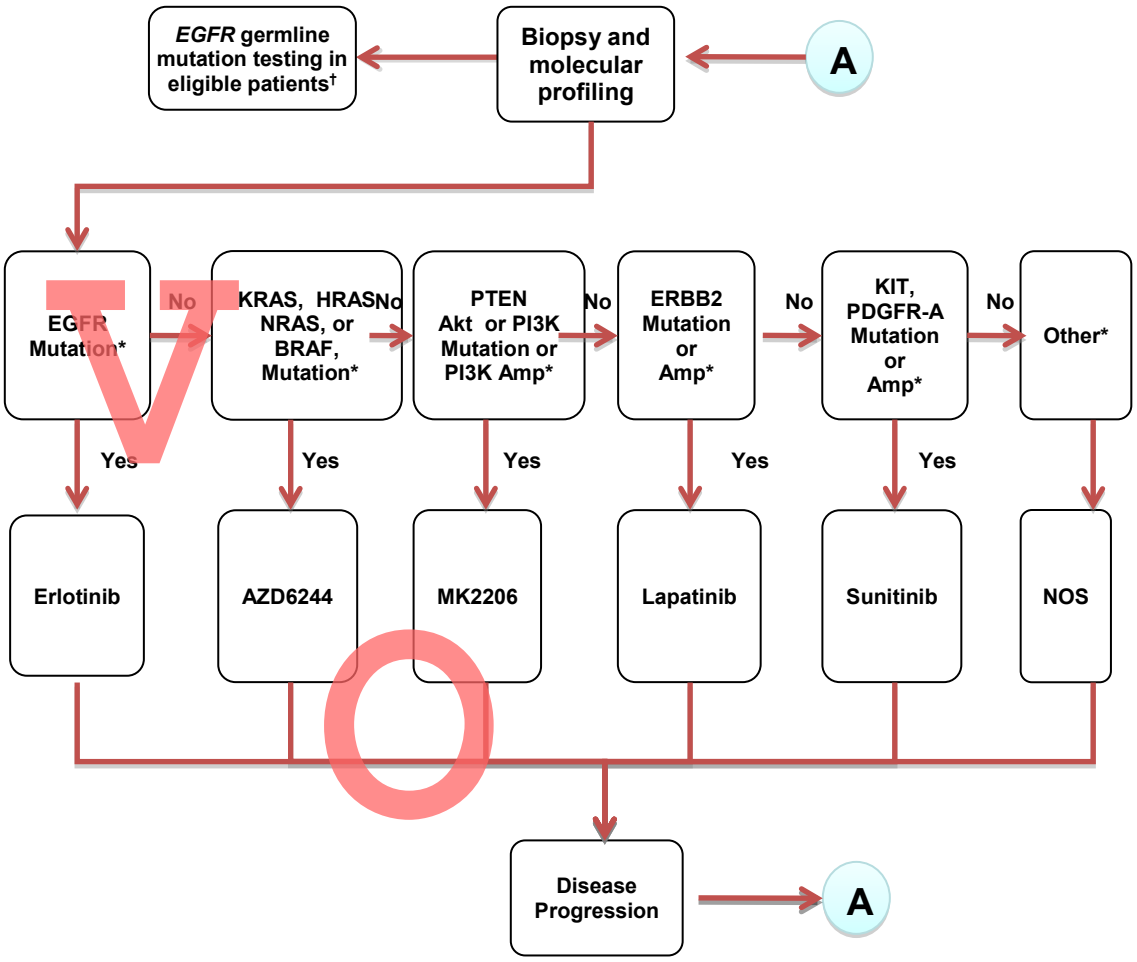
5.3 Enrollment in treatment arms

Treatment arms are no longer recruiting as of 3/22/16.

Due to the rapidly progressing course of the thoracic malignancies in this study, treatment decisions will be made in approximately 2 weeks from the time that the tumor samples were obtained. Based on the molecular profiling results, the patients will be offered enrollment into a specific treatment arm according to the decision tree shown in Figure 2 and the eligibility criteria described in Section 3.

Individuals who meet the eligibility criteria for *EGFR* germline mutation testing (Section 3.4) but who do not have advanced cancer as defined in Section 3.1.1 are not eligible for the treatment arms.

Figure 2. Schema



*** See Sections 3.2 and 3.3 for eligibility criteria for the treatment arms. Patients will be allowed to crossover to different treatment arms as long as they meet eligibility criteria.**

† See separate eligibility criteria (Section 3.4) and schema for *EGFR* germline mutation testing.

No = No or not available

Amp = gene amplification by FISH

NOS = non-otherwise specified arm

A = optional re-biopsy for patients that responded to previous line of therapy

Each study arm is independent from the other arms; it has its own eligibility criteria (Section 3) and statistical considerations (Section 15), and will be analyzed independently. Additionally, it is not our intention to compare the outcomes among treatment arms since the patients enrolled in each arm will have different molecular characteristics.

Patients enrolled in one of the treatment arms of this protocol, with the exception of the NOS arm and individuals enrolled for *EGFR* germline mutation testing only, will be treated and

followed with clinical visits at least every 3, 4, or 6 weeks, as defined for each treatment arm; i.e., every 3 weeks for the Erlotinib, AZD6244, and Lapatinib arms; every 4 weeks for the MK-2206 arm; and every 6 weeks for the Sunitinib arm. The time between clinic visits may be extended at the PI's discretion by up to 1 full cycle, for a maximum of 2 cycles between clinic visits (i.e., clinic visits may be extended to every 6 weeks for the Erlotinib, AZD6244, and Lapatinib arms; every 8 weeks for the MK-2206 arm; and every 12 weeks for the Sunitinib arm). If the frequency of clinic visits is extended, laboratory tests will not be required nor samples collected until the next clinic visit. Re-staging imaging will be performed at the end of every two cycles as defined for each treatment arm; i.e., every 6, 8, or 12 weeks +/- 1 week. See Section 5.9.2 for details. The patients will continue on the selected arms until disease progression.

5.4 Treatment after disease progression

Patients who initially had a response to treatment or a prolongued stabilization of the tumor (>6 months) but that eventually develop progressive disease, will be offered the option to have a repeat biopsy for molecular profiling analyses. This biopsy will have the intention to study potential mechanisms of resistance to specific targeted therapies. This will also be potentially used for treatment decisions or enrollment in other molecularly targeted protocols. However, if the patient refuses this option, we will use the most recent results of the molecular profiling analyses to verify eligibility for enrollment in an alternative treatment arm on this study (crossover).

At the time of disease progression, the patients will be allowed to re-register onto another treatment arm that is available in this protocol, according to the schema, and as long as they still meet the eligibility criteria described in Section 3. In this case, patients will be re-registered onto the treatment arm, but not re-consented. The patient will count as a new patient to that specific arm, keeping their original patient ID number.

5.5 The NOS arm

All patients that undergo molecular profiling but that are not enrolled into any of the investigational treatment arms will be enrolled into the NOS arm, which is considered to be a natural history arm. These patients do not have to meet any particular eligibility criteria. Individuals who meet the eligibility criteria for *EGFR* germline mutation testing (Section 3.4) but who do not have advanced cancer as defined in Section 3.1.1 are not eligible for the NOS arm.

This will allow data collection on these patients, linking their clinical information with their molecular profiling analyses. Frequency determination of each mutation, its association with clinical features, response to other standard and experimental treatments and overall outcome data will be collected. As future therapeutic protocols or treatment arms specific for their molecular profiles are developed at NIH or elsewhere, patients may be notified of their eligibility for these studies.

The NOS arm will consist of the following mixture of patients:

- Patients that do not meet the eligibility criteria for enrollment in the available treatment arms of this protocol.
- Patients that meet eligibility criteria for a particular arm but cannot be enrolled on it because of other reasons such as patient preference or drug availability issues, among others.

The NOS arm is considered a natural history arm in which the patients are allowed to receive a standard of care or experimental therapy of their choice, including but not limited to the following modalities:

- Patients will be allowed to participate on any available treatment clinical trial at NIH or elsewhere.
- Patients will be allowed to receive standard of care treatment at NIH under protocol 04-C-0165 ("Care of the Adult Oncology Patient, Center for Cancer Research, NCI;" Principal Investigator: James Gulley, M.D.) or elsewhere.
- Patients will be allowed to receive best supportive care at NIH or elsewhere.

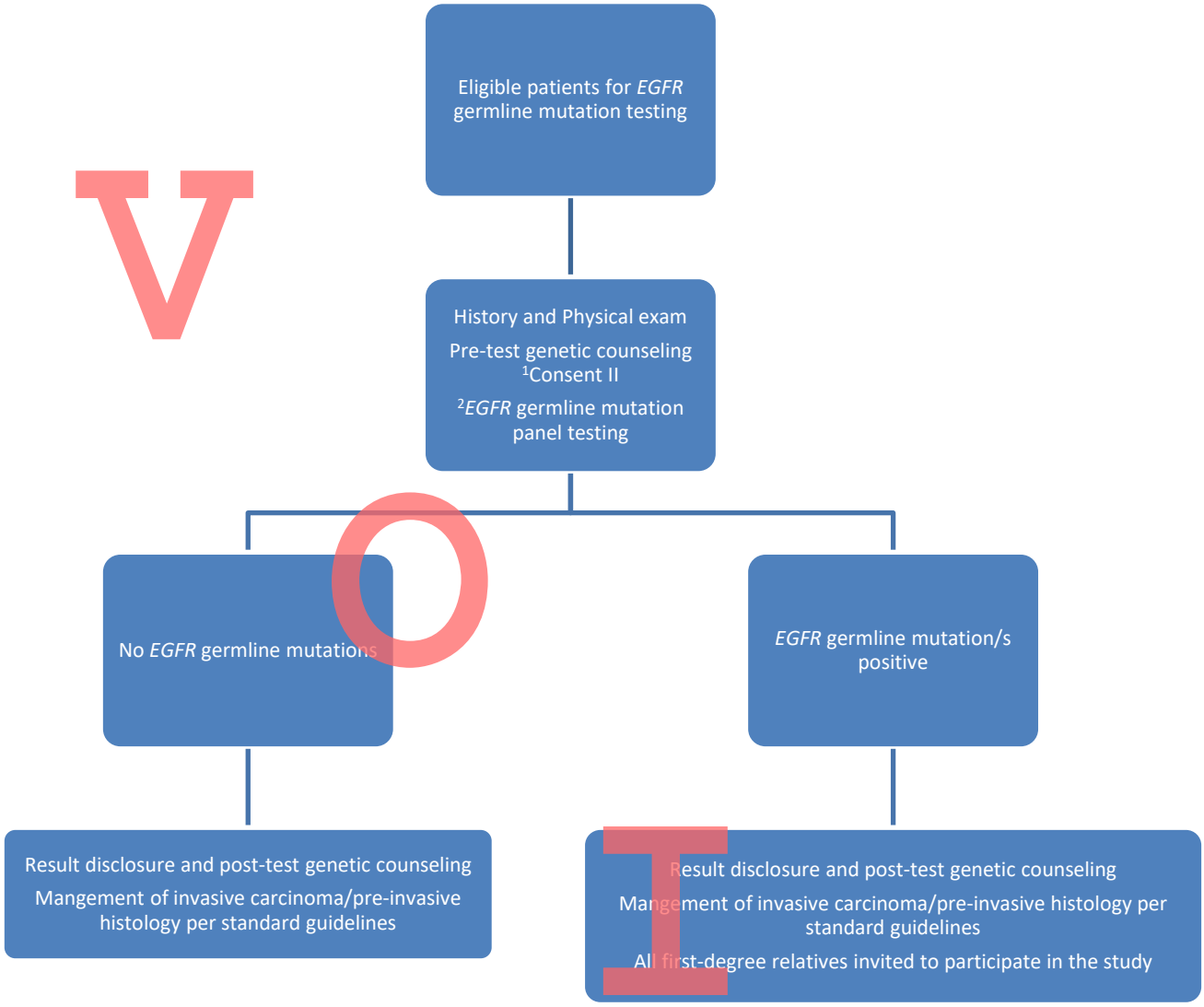
Patients on this arm will be followed either with clinic visits, phone interviews, or medical records review every 4 months (+/- 2 months). The patients will continue to be followed in this arm indefinitely, until death or until the patient decides to withdraw from the protocol.

5.6 *EGFR* germline mutation testing

Patients at the NCI site meeting the eligibility criteria in Section **3.4** will be enrolled in *EGFR* germline mutation arm of the study (100, 101). Guidelines set forth by American Society of Clinical Oncology (ASCO) in 2003 and updated in 2010 will be followed in obtaining consent, genetic counseling, testing and follow up (86, 102).

Evaluation and follow-up of eligible patients and family members are outlined in Figure 3 and Figure 4.

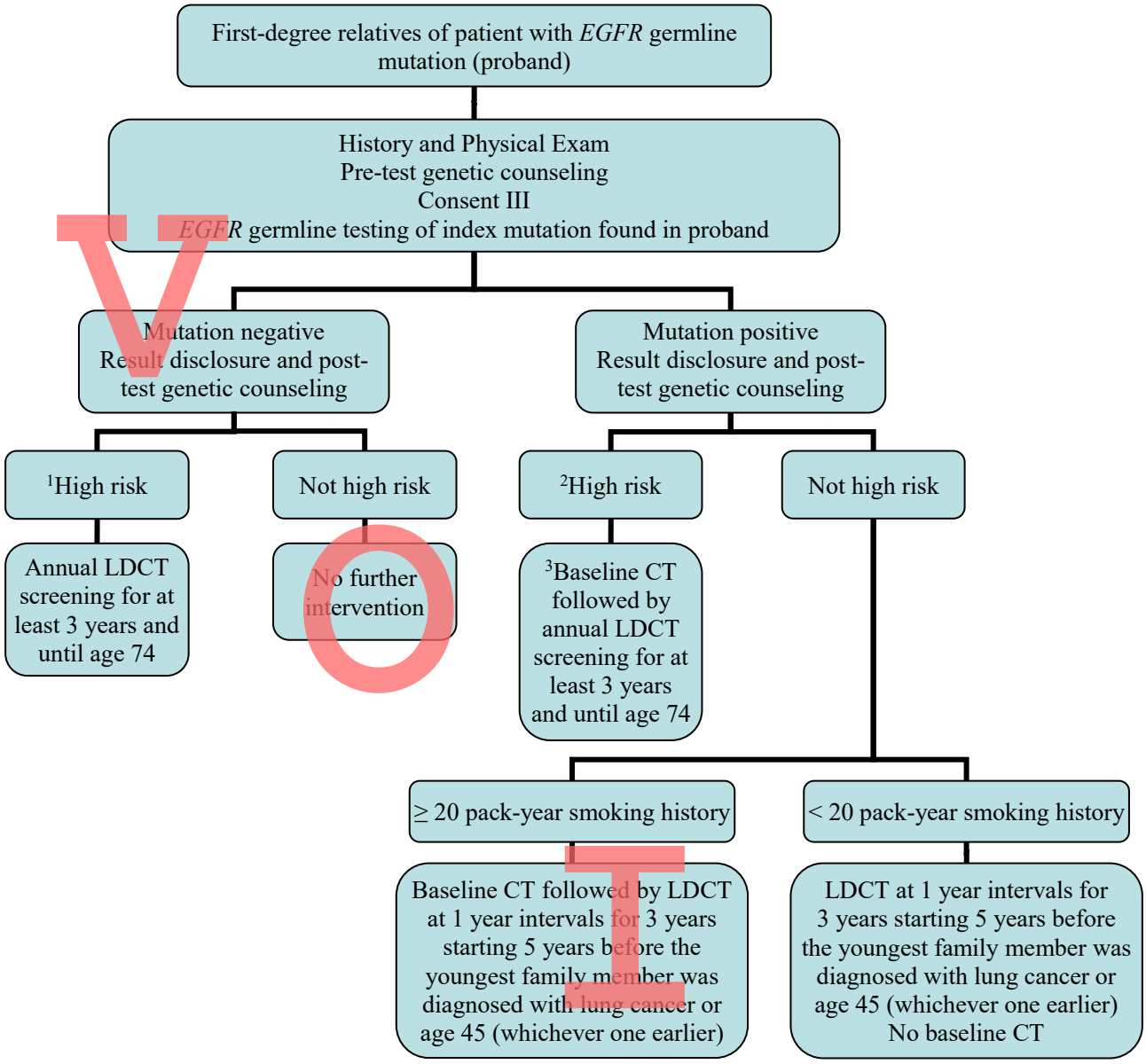
Figure 3. Flow chart depicting evaluation and follow-up of eligible patients who undergo *EGFR* germline mutation testing.



[1] Consent II: Informed consent for testing for *EGFR* germline mutations.

[2] *EGFR* germline mutation panel: *EGFR* exon 20 T790M, exon 21 V843I, exon 21 R831C and exon 20 R776G.

Figure 4. Flow chart depicting evaluation and follow-up of first-degree relatives who undergo *EGFR* germline mutation testing.



[1] Between 55 and 74 years of age at enrollment with a history of cigarette smoking of at least 30 pack-years, and, if former smokers, those who quit within the previous 15 years.

[2] Age ≥ 45 years at enrollment with ≥ 20 pack-year smoking history or age within 5 years before the youngest family member was diagnosed with lung cancer.

[3] For lung nodules and incidental lesions detected on CT, follow up is per standard guidelines.

Consent III: Informed consent for testing for *EGFR* germline mutations in first-degree relatives of individuals with known *EGFR* germline mutations.

Abbreviation: LDCT, low-dose screening computed tomography scan.

Follow-up of lung nodules recommended by Fleischner Society (1) and National Comprehensive Cancer Network(2)(2)(2).

5.6.1 Consent

The proband as well as all family members undergoing germline genetic testing will sign informed consent after undergoing genetic counseling and prior to any germline genetic testing. Consents I, II and III will be used as follows:
Consent I: Patients who are eligible for the molecular profiling protocol will sign the standard consent for molecular profiling protocol.

Consent II: Individuals with lung cancer or a pre-invasive histology who meet the eligibility criteria for *EGFR* germline mutation testing (i.e., meets criteria **3.4.1** and **3.4.2**) will sign consent II (i.e., consent for germline mutation testing of proband). Consent II will be obtained from the next of kin for deceased family members of the proband with a history of lung cancer for whom tissue is available.

Consent III: First-degree relatives of individuals with *EGFR* germline mutations (with no known lung cancer and/or germline mutations, i.e., meets criteria **3.4.1** and **3.4.3**) will sign consent III (i.e., consent for germline mutation testing of unaffected family member).

5.6.2 Pre-test genetic counseling

Genetic counseling will be provided for the individual and family members both before and after germline mutation analysis as detailed below: The proband and family members who undergo genetic testing will meet a genetics healthcare provider (GHP) (either in clinic or via phone conference) prior to signing consent for germline testing and undergoing germline genetic testing. The GHP will explain the specific testing to be performed, the timeframe for analysis, follow-up testing that may be required, and the types of information that may be obtained and potential healthcare recommendations which could result based on potential results. Ample time will be given to ensure that all questions are answered. At each step in the process the GHP will be available to answer questions and assist the individual in the decision process regarding testing and results.

5.6.3 Genetic testing

Step 1: The proband (patient who presents to our clinic with lung cancer or a pre-invasive histology with more than two affected family members with invasive lung cancer or one of the pre-invasive histologies associated with the development of lung cancer) will be tested for *EGFR* germline mutations (on peripheral blood mononuclear cells) that have been associated with familial clustering of lung cancer (*EGFR* exon 20 T790M, exon 21 V843I, exon 21 R831C and exon 20 R776G). The following information will be documented for the proband: mutational status of the tumor, treatment history and response to EGFR TKI, age, sex, ethnicity, detailed family history with documentation of any malignencies in the family, smoking survey based on the Centers for Disease Control and Prevention Behavioral Risk Factor Surveillance System Survey Questionnaire (**Appendix K: Tobbacco Use History Questionnaire**), environmental exposure and previous malignancies. Once a specific mutation is confirmed in the proband, all adult first-degree relatives (parents, offspring, and siblings) will be invited to participate in this study.

Step 2: Recruitment of first-degree relatives

If the proband consents to do so, all first-degree relatives (of the proband) will be contacted by a letter followed by phone call.

Step 3: Consenting family members of the proband will be asked to undergo germline mutation testing of the index mutation, which was identified in the proband. For all family members, a

history and physical exam will be performed and the following information documented: age, sex, ethnicity, detailed family history with documentation of any malignencies, smoking survey based on the Centers for Disease Control and Prevention Behavioral Health Factor Surveillance System Survey Questionnaire (**Appendix K: Tobbacco Use History Questionnaire**), environmental exposure and previous malignancies.

In many cases, due to the high case-fatality rate of lung cancer, the family member with lung cancer may be deceased. In these cases, archived FFPE tissue may be used for germline mutation testing (after genetic counseling and obtaining consent from next of kin), if normal tissue can be isolated using laser capture microdissection.

5.6.4 Result disclosure and post-test counseling

Results of *EGFR* germline mutation testing will be available in Clinical Research Information System (CRIS) in ≤ 2 weeks after date of sample collection. Disclosure of test results will be performed by a study investigator and GHP, who will provide any management recommendations as well as recommendations for testing family members as applicable. Study participants and their designated local health care provider(s) will also receive these recommendations in writing as a counseling letter.

5.6.5 Follow-up

The follow up of individuals who undergo germline mutation testing will follow the schema in Figure 3. After result disclosure and post-test counseling, whether an *EGFR* germline mutation is found or not, mangement of invasive carcinoma/pre-invasive histology will follow standard guidelines.

The follow up of first-degree relatives of the proband who undergo germline mutation testing will follow the schema in Figure 4.

Mutation negative individual: These individuals will be will be risk stratified based on smoking history into "high-risk" (individuals between 55 and 74 years of age at enrollment with a history of cigarette smoking of at least 30 pack-years, and, if former smokers, those who quit within the previous 15 years) and "not high-risk" (individuals not meeting the high-risk criteria) groups (103). In the high-risk group, annual low-dose computed tomography (LDCT) screening (CT screening techniques will follow NCCN guidelines; an example of the latest guideline is in Appendix L) will be rcommended for at least 3 years and until age 74 (103). No further intervention will be recommended for the not high-risk group.

Mutation positive individual: These individuals will be will be risk stratified based on smoking history and age of youngest family member with lung cancer into "high-risk" (age ≥ 45 at enrollment with ≥ 20 pack-year smoking history or age within 5 years before the youngest family member was diagnosed with lung cancer) and "not high-risk" (individuals not meeting the high-risk criteria) groups. In the high-risk group, annual LDCT screening will be rcommended for at least 3 years and until age 74 (103). The not-high risk individuals will be further risk-stratified based on smoking history into 2 groups and followed as described: ≥ 20 pack-year history of smoking [Baseline CT followed by LDCT at 1 year intervals for 3 years starting 5 years before the youngest family member was diagnosed with lung cancer or age 45 (whichever one earlier)] and ≤20 pack year history of smoking [LDCT at 1 year intervals for 3 years starting 5 years before the youngest family member was diagnosed with lung cancer or age 45 (whichever one earlier)].

All probands as well as family members who are enrolled in the protocol will be counseled regarding smoking cessation and will be offered pharmacologic assistance if needed at NIH. All consented individuals (probands and first-degree relatives) will remain on study until death or withdrawal of consent to participate. Survival data, adherence to screening recommendations and details of incident cancers (date of diagnosis, stage at diagnosis) will be documented in first-degree relatives using a phone call once a year. The proband will be followed using a phone call once a year to collect survival data.

5.6.6 Laboratory and specimen information

Peripheral blood samples (3-5 mL) will be submitted in sodium citrate anticoagulated "light blue top" tubes to the Molecular Diagnostics Laboratory of the Laboratory of Pathology, NCI for analysis of the following germline mutations affecting the *EGFR* gene: exon 20 T790M, exon 21 V843I, exon 21 R831C and exon 20 R776G. DNA extraction is performed on a PSS Biosystems (Pleasanton, CA) robotic device using the company's proprietary bead-based DNA extraction kit. Targeted analysis for potential germline *EGFR* mutations will be performed by pyrosequencing on a PyroMark Q24 instrument (Qiagen, Valencia, CA) after initial PCR of the targeted regions. Primers for all PCR and pyrosequencing reactions are designed and validated by the Molecular Diagnostics Laboratory (MDU) of the Laboratory of Pathology, Center for Cancer Research, NCI, NIH, and are available upon request. The MDU is a CLIA-regulated and CAP-inspected high-complexity clinical molecular diagnostics laboratory (CLIA# 21D0716664; Exp. 7/16/2012). The report will be issued within 2 weeks from the date of collection and will be available in Clinical Research Information System (CRIS).

5.6.7 Data collection

A full pedigree will be collected on each family enrolled in this aspect of the study. This study will utilize the genetic database Progeny for collection and storage of pedigree data. Progeny is an online multi-user clinical data management system used to track family history, phenotype, and genotype data. The Progeny database is stored on a secure NCI server that is password protected. The database itself is also password protected with the options to restrict each class of users to specifically defined access and read/write privileges based on their role on the study. The database also has the capability to track passwords, logins, previous entry, new entry and user information with a date/time stamp. Families and individual subjects within families are tracked by distinct identification labels that include a unique numeric family and individual identifier.

5.6.8 Description of how privacy and confidentiality of medical information/biological specimens will be maximized

Biospecimens will be collected and processed using validated SOPs that will ensure both specimen quality and patient confidentiality pursuant to informed consent provisions.

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. Coded, linked labels 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 de-linked specimens will be shipped for analysis and/or storage. Coded, linked specimen labels will indicate: protocol number, order in which the patient was enrolled in the trial, type of sample, collection time, and total volume collected, as appropriate.

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 personnel only. The only patient information available in the inventory system will be the patient sex, diagnosis, and level of informed consent provided. SOPs ensure that any changes in the 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.

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).

The PI will record any loss or unanticipated destruction of samples as a deviation. Reports will be made per the requirements of section **8.2**.

A Certificate of Confidentiality will be obtained from the Department of Health and Human Services before germline mutation testing begins. The investigators will use the Certificate to protect against the compelled disclosure of personally identifiable information and to support and defend the authority of the Certificate against legal challenges. Personnel involved in the conduct of the research will comply with all the requirements of 45 CFR Part 46 "Protection of Human Subjects." All research participants will be informed that a Certificate has been issued, and they will be given a description of the protection provided by the Certificate.

5.7 Agent Administration

Treatment will be administered on an outpatient basis. Reported adverse events and potential risks from the agents used in this protocol are described in Section **7**. Appropriate dose modifications for these agents are described in Section 6. No investigational or commercial agents or therapies other than those described below may be administered with the intent to treat the patient's malignancy.

Patients in all treatment arms (Erlotinib, AZD6244, MK2206, Lapatinib and Sunitinib) will be provided with a Medication Diary (**Appendix B**). They will be instructed in its use and asked to bring the diary with them to each appointment. A new copy of the Medication Diary will be given to patients whose dose is reduced due to adverse events. Patients should log in their daily dose onto the diary, including missed, skipped, or vomited doses. If the patient vomits after taking the tablets, the dose is replaced only if the tablets can actually be seen and counted. If a patient misses a dose, he or she should be instructed to resume dosing with the next scheduled dose.

5.7.1 CTEP agents

5.7.1.1 Erlotinib

1) Erlotinib will be administered at a dose of 150 mg once daily every three weeks.

2) Tablets should be taken once daily preferably in the morning with up to 200 mL of water 1 hour before or 2 hours after food.
3) Patients should wear sunscreen protection, hat, and long sleeves to avoid sun as it can exacerbate skin rash.
4) Patients should be instructed to store Erlotinib at room temperature.
5) Patients should be advised to drink plenty of water or take rehydration fluids to avoid dehydration if diarrhea occurs.

5.7.1.2 AZD6244 Hydrogen Sulfate

1) AZD6244 hydrogen sulfate will be administered at a dose of 75 mg twice daily (BID), approximately 12 hours apart, every three weeks.
2) Advise the patient to take AZD6244 doses on an empty stomach (no food or drink other than water for 1 hour before or 2 hours after dosing).
3) Patients should be instructed to store AZD6244 at room temperature.
4) Patients should be advised to drink plenty of water or take rehydration fluids to avoid dehydration if diarrhea occurs.
5) Patients should avoid excessive sun exposure and use adequate sunscreen protection if sun exposure is anticipated.

5.7.1.3 MK-2206

1) MK-2206 will be administered orally at a dose of 200 mg weekly every four weeks.
2) Patients should take MK-2206 - 2 hours before or after a meal.
3) Patients should be instructed to store MK-2206 at room temperature.
4) Patients should be advised to drink plenty of water or take rehydration fluids to avoid dehydration if diarrhea occurs.

5.7.1.4 Lapatinib

1) Lapatinib will be administered orally at a dose of 1500 mg daily every three weeks.
2) Patients should take Lapatinib - 1 hour before or after a meal.
3) Patients should be instructed to store Lapatinib at room temperature.
4) Patients should be advised to drink plenty of water or take rehydration fluids to avoid dehydration if diarrhea occurs.
5) Routine monitoring for cardiac function (ECHO/MUGA) should be performed at baseline and every other cycle of treatment.

5.7.2 Other agents

5.7.2.1 Sunitinib

1) Sunitinib will be administered orally at a dose of 50 mg daily for 4 consecutive weeks followed by 2 weeks of rest (drug holiday) with no sunitinib every 6 weeks.

2) Patients will take sunitinib once daily in the morning, with or without food, as desired.
3) Patients should be instructed to store Sunitinib at room temperature.
4) Patients should be advised to drink plenty of water or take rehydration fluids to avoid dehydration if diarrhea occurs.
5) Because hypertension is a known and potentially serious but rare adverse event associated with sunitinib malate treatment, patients will have their blood pressure monitored and recorded weekly during the first cycle of therapy, either at the doctor's office or using any calibrated electronic device (such as those found at a local drug store or pharmacy). (See Section 6 for hypertension management and dose reduction guidelines.)
6) Routine monitoring for cardiac function (ECHO/MUGA) should be performed at baseline and then every other cycle of treatment in the following groups of patients: (1) those entering the trial with NYHA Class II cardiac dysfunction (see Appendix D), (2) those with a history of Class II heart failure who are asymptomatic on treatment, and (3) in those previously exposed to anthracyclines or thoracic irradiation if the heart was included in the radiotherapy port. Routine cardiac monitoring is not required in patients with no known cardiac dysfunction at entry or in the absence of clinically observed adverse cardiac events.
7) Although adrenal gland insufficiency is rarely seen with sunitinib treatment, patients should be clinically followed for the signs and symptoms of this complication, especially (1) patients with comorbidities associated with adrenal dysfunction, (2) patients with pre-existing adrenal insufficiency (primary or secondary), and (3) patients with concomitant stress (e.g., fever, infection, bleeding, serious accident, surgery) that may precipitate overt adrenal insufficiency in the presence of subclinical sunitinib-induced adrenal toxicity. If clinically indicated, objective testing for adrenal gland function should be conducted.
8) Patients with bulky solid tumors should be monitored closely for pneumothorax, intestinal fistulae, or intestinal perforation in the event of rapid tumor destruction.
9) Patients should be alerted to the possibility that sunitinib capsules can cause a yellow discoloration of the skin on direct contact. If this happens, the patient should wash immediately with soap and water.

5.8 General Concomitant Medication and Supportive Care Guidelines

5.8.1 General Guidelines: Concomitant Medications

A concomitant medication is any medication a patient entering the trial is using and is expected to continue using for some portion of the trial as well as any medication the patient uses during the course of the trial. Study drugs are not considered concomitant medication.

All concomitant medications recorded at trial entry must have a related, ongoing concomitant illness listed under the medical history at the time of patient entry into the trial unless the medication is used for prophylaxis. Patients may continue to use any ongoing medications not prohibited by the protocol. All prescription and over-the-counter medications at trial entry as well as any new medications started during the trial must be documented. The documentation should continue until 30 days from the end of the last study drug treatment.

No other anti-cancer therapy including chemotherapy, radiation therapy, hormonal cancer therapy and immunotherapy, or experimental medications are permitted while the patients are receiving treatment under one of the targeted therapy arms of this trial. Any disease progression that requires other specific anti-tumor therapy will be cause for discontinuation from study medication. Patients who are receiving steroids for complications of their primary malignancy may enroll on the study and continue receiving steroids as long as needed. Because there is a potential for interaction of all of the medications used in the experimental arms, with other concomitantly administered drugs through the cytochrome P450 system, the case report form (CRF) must capture the concurrent use of all other drugs, over-the-counter medications, or alternative therapies. The Principal Investigator should be alerted if the patient is taking any agent known to affect or with the potential to affect selected CYP450 isoenzymes. A comprehensive list of CYP3A4 inhibitors, inducers, and substrates is provided (see **Appendix F**). In addition, patients and their caregivers should be provided the patient information sheet (see **Appendix G**) describing potential interactions of the study agents with other drugs, remedies, and medications.

Additionally, the patient's smoking history should be documented in the CRF, including the number of packs smoked/day (if a current smoker).

5.8.1.1 Diarrhea

Diarrhea should be treated with the following regimen: loperamide (4 mg PO) at onset of symptoms, followed by 2 mg loperamide every 2 hours while awake (or 4 mg PO every 4 hours while sleeping) up to a maximum of 16 mg loperamide per day.

5.8.1.2 Nausea and vomiting

The nausea and vomiting that may occur with investigational agents should be managed through the use of appropriate simple supportive measures (e.g., prochlorperazine or metoclopramide).

5.8.1.3 Hand-foot syndrome

This may be treated with topical emollients (such as Aquaphor), topical or systemic steroids, and/or antihistamine agents. Vitamin B6 (pyridoxine; 50-150 mg orally each day) may also be used. Avoid exposure to heat, hot water, pressure, or friction. Use of soft, well-fitting shoes may help, as may use of acetaminophen if needed for analgesia.

5.8.1.4 Rash

Rash should be treated with standard acne therapies, including topical and oral antibiotics used to treat acne such as the following: minocycline, topical tetracycline, topical clindamycin, topical silver sulfadiazine, diphenhydramine, or oral prednisone (short course).

5.8.1.5 Other protective / preventative measures

Patients on Erlotinib should wear sunscreen protection, hat, and long sleeves to avoid sun as it can exacerbate skin rash.

5.8.1.5.1 Myelossupression related complications

Patients with neutropenic fever or infection should be evaluated promptly and treated with IV antibiotic therapy or therapeutic colony-stimulating factors as appropriate

following the ASCO guidelines [J Clin Oncol 18(20):3558-85, 2000]. Packed red blood cell and platelet transfusion should be administered as clinically indicated. Erythropoietic agents may be used at the discretion of the treating physician.

5.8.1.5.2 Additional recommendations for patients in Sunitinib arm

- Use of agents with proarrhythmic potential (terfenadine, quinidine, procainamide, disopyramide, sotalol, probucol, bepridil, haloperidol, risperidone, indapamide, and flecainide) is not permitted during treatment with this drug. A comprehensive list of agents with proarrhythmic potential can be found at http://torsade.org .
- Sunitinib is primarily metabolized by liver enzymes, particularly CYP3A4. Co-administration of potent inhibitors or inducers of this enzyme can result in significant changes in exposure to sunitinib (e.g., a mean 1.8-fold increased exposure with ketoconazole and a mean 4-fold decrease with rifampin). For this reason, use of the following agents is not permitted before or during enrollment in this arm of the study:

Inhibitors – prohibited 7 days before dosing and during study.

azole antifungals (ketoconazole, itraconazole)	Verapamil
clarithromycin	HIV protease inhibitors (indinavir, saquinavir, ritonavir, atazanavir, nelfinavir)
erythromycin	Delavirdine
diltiazem	

Inducers – prohibited 12 days before dosing and during study.

rifampin	Phenytoin
rifabutin	St. John's wort
carbamazepine	Efavirenz
phenobarbital	Tipranavir

- Steroid use is not recommended during sunitinib treatment unless absolutely necessary (e.g., for treatment of adverse events or protocol-required premedication) because many steroids (e.g., prednisone, prednisolone, dexamethasone, etc.) effectively lower sunitinib exposure through CYP3A4 interactions.
- The use of coumarin-derivative anticoagulants such as warfarin (Coumadin®) is not recommended, although doses of up to 2 mg daily are permitted for prophylaxis of thrombosis.

5.8.1.5.3 Additional recommendations for patients in AZD6244 arm

- Ophthalmologic exam (including slit-lamp) – to be performed in patients experiencing visual disturbances while in the trial.

- Echocardiogram or MUGA scan (measurement of LVEF) – to be performed in patients with symptoms (AEs) suggestive of cardiac impairment (congestive heart failure, oedema, dyspnea).
- Decreases in LVEF from baseline (if known) may be investigated according to the algorithm (note- this appears as a word doc in the in AZD6244 ISS study guide)
- All new dyspnea AEs or worsening of pre-existing dyspnea AEs should be followed according to the dyspnea algorithm (note this appears in the AZD6244 ISS study guide)

5.9 Protocol Evaluation

5.9.1 Prior to each treatment cycle (please refer to Section **5.3** for definition of a cycle):

5.9.1.1 SOAP progress note.

5.9.1.2 Laboratory examination:

5.9.1.2.1 Hematological Profile: CBC with differential and platelet count.

5.9.1.2.2 Biochemical Profile: Sodium, potassium, chloride, CO2, BUN, creatinine, glucose, AST, ALT, alkaline phosphatase, bilirubin, albumin, total protein, LDH, calcium, phosphorous and magnesium.

5.9.1.3 Vital signs including weight.

5.9.2 Restaging and Imaging:

Restaging will be performed at the end of every two cycles, +/- 1 week, as defined for each treatment arm. Therefore, restaging will be performed every 6 weeks +/- 1 week for the Erlotinib, AZD6244, and Lapatinib arms; every 8 weeks +/- 1 week for the MK-2206 arm, and every 12 weeks +/- 1 week for the Sunitinib arm. Imaging modalities will consist of CT or MRI, as appropriate for disease evaluation. The use of FDG-PET will be allowed but it would be considered exploratory and its results will not be used for treatment decisions.

5.10 Duration of therapy / off treatment criteria

In the absence of treatment delays due to adverse events, treatment under any of the investigational arms (Erlotinib, AZD6244, MK2206, Lapatinib and Sunitinib) may continue until one of the following criteria applies:

- Disease progression as defined by Section **13**.
- Intercurrent illness that prevents further administration of treatment.
- Delay of treatment due to toxicity of ≥3 weeks.
- Unacceptable adverse event(s)*.
- Non-compliance to therapy regimen.
- Pregnancy
- Patient decides to withdraw from the study arm or stop therapy.

- Deterioration of the patient's condition that render further treatment unacceptable in the judgment of the investigator.

* Patients removed from study arm due to unacceptable adverse events, will continue to be followed in clinic, at least every 4 weeks until all adverse events have resolve to grade I or less.

5.11 Duration of follow up

All patients that are not receiving active investigational treatment (Erlotinib, AZD6244, MK2206, Lapatinib and Sunitinib), including those on the NOS arm and those deemed as "Screen Failures," will continue to be followed until death or the patient decides to withdraw from the study. These patients will be followed either with clinic visits or phone interviews every 4 months (+/- 2 months). If patient is receiving treatment at an outside facility, pertinent medical records should be sent to the PI at the participating institution for data collection. The following information will be collected:

- Date of follow up
- Is patient dead or alive?
- If dead, document exact date of death.
- Further chemotherapy / biotherapy treatment(s)
    - Drug name(s)
    - Date(s) of administration (month/year)
    - Response to treatment
- Document date of disease progression
- Document ECOG PS

* No toxicity information will be collected for patients being followed in the NOS arm.

5.12 Off Study Criteria

Patients will be removed from study for:

- Screening failure
- Death
- Patient decides to withdraw from the study
- At the investigator's discretion
- Lost to follow-up

The reason for study removal and the date the patient was removed will be documented in the Case Report Form.

5.13 Off-Study Procedure

Authorized staff must notify Central Registration Office (CRO) when a subject is taken off-study. A Participant Status Updates Form from the web site (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.

6 DOSING DELAYS/DOSE MODIFICATIONS

Patients enrolled in one of the experimental study arms may continue to receive additional cycles of therapy provided that the patient meets the following criteria on Day 1 of each cycle:

- ANC > 1,000/mcL
- Platelets > 100,000/mcL
- Non-hematologic toxicity recovered to < grade 2 (or tolerable grade 2 or baseline)
- No evidence of progressive disease

In the event of an adverse event at least possibly related to the agent, the doses of such agent should be adjusted according to the guidelines shown in the Dose Delays / Dose Modifications tables shown below (Sections **6.1** and **6.2**). If an adverse event is not covered in such tables, doses may be reduced or held at the discretion of the investigator for the subject's safety.

Subjects with adverse events that are manageable with supportive therapy may not require dose reductions (e.g., nausea/vomiting may be treated with antiemetics, diarrhea may be treated with loperamide, and electrolyte abnormalities may be corrected with supplements rather than by dose reduction).

Subjects will be withdrawn from the study arm if they fail to recover to CTC Grade 0-1 or tolerable grade 2 (or within 1 grade of starting values for pre-existing laboratory abnormalities) from a treatment-related adverse event within 21 days OR they experience agent related adverse events requiring dose modification despite two previous dose reductions (i.e. would require a 3rd dose reduction) unless the investigator and CTEP monitor agree that the subject should remain in the study arm because of evidence that the patient is/may continue deriving benefit from continuing study treatment (i.e. patient has PR, CR, SD > 3 months). The appropriate reduced dose will be determined after discussion between the principal investigator and CTEP monitor.

6.1 CTEP agents

6.1.1 Erlotinib

Table 6. Erlotinib Monotherapy Dose Level Reductions

Dose Level	**Erlotinib Dose**
Starting Dose	150 mg/day
First Dose Reduction	100 mg/day
Second Dose Reduction	75 mg/day

Table 7. Dosage Modification Criteria for **Erlotinib**–related Toxicities

Toxicity	**Grade**	**Erlotinib dosage modification***	**Guideline for management**
Keratitis	1	None	No intervention
	2 (if < 14 days)	None	Preservative-free artificial tears, ointments, and/or other therapies as clinically indicated, with a follow-up examination within 2 weeks
	2 (if >14 days)	Hold until recovery to ≤ grade 1 And then Reduce 1 dose level	
	≥3	Hold until recovery to ≤ grade 1 And then Reduce 1 dose level*	
Diarrhea	1	None	No intervention
	2	None if tolerable If not tolerable, hold until resolution to grade ≤ 1, then resume at the same dose. If intolerable grade 2 recurs after resumption, hold until resolution to grade ≤ 1 and resume with 1 dose reduction.	Loperamide (4 mg at first onset, followed by 2 mg every 2–4 hrs until diarrhea free for 12 hrs)
	≥ 3 (despite optimal loperamide use)	Hold until recovery to ≤ grade 1 And then Reduce 1 dose level*	
Rash	1	None	No intervention
	2	None if tolerable If not tolerable, hold until resolution to grade ≤ 1, then resume at the same dose. If intolerable grade 2 recurs after resumption, hold until resolution to grade ≤ 1 and resume with 1 dose reduction.	Any of the following: minocycline, topical tetracycline or clindamycin, topical silver sulfadiazine, diphenhydramine, oral prednisone (short course)
	≥ 3	Hold until recovery to ≤ grade 1 And then Reduce 1 dose level	

Bilirubin	≥3 x ULN	Hold until grade ≤2 And then Reduce 1 dose level	
Liver transamin ase	> 5 x ULN	Hold until grade ≤2 And then Reduce 1 dose level	
Signs and symptoms of interstitial pneumonitis		Hold pending diagnosis Permanently discontinue if diagnosis is confirmed and considered possibly related to erlotinib	Patient should be thoroughly evaluated, closely monitored, and supported as clinically indicated
Other toxicity	≥ 2 prolonged clinically significant toxicity	Hold until recovery to ≤ grade 1 And then Reduce 1 dose level*	Treatment as appropriate
*if dose has been previously held for grade 2 rash or diarrhea, and grade 2 symptoms recur, OR if the patient finds the symptoms unacceptable, hold dose until recovery to ≤ grade 1 and then reduce dose one level			

6.1.2 AZD6244 Hyd-Sulfate

Table 8: AZD6244 Monotherapy Dose Level Reductions

Dose Level	**AZD6244 Dose**
Starting Dose	75 mg BID
First Dose Reduction	50 mg BID
Second Dose Reduction	50 mg once daily

Table 9: Dosage Modification Criteria for AZD6244–related Toxicities

Event	**AE Grade or Observation**	**Dose modification**
Dermatology/Skin	Grade 1 or 2	Maintain dose
	Grade 3 or 4 or intolerable grade 2	Hold AZD6244 until < tolerable grade 2, then reduce 1 dose level and resume treatment [1].
	Recurrent Grade 3	Hold AZD6244 until < tolerable grade 2, then reduce 1 additional dose level and resume treatment [1].

	Recurrent Grade 3 after 2 dose reductions	Remove patient from study arm.
	Grade 4	Patients with grade 4 rash (i.e. life threatening generalized exfoliative ulceration; erythemia multiforme/toxic epidermal necrolysis should be removed from study arm.
Diarrhea (if anti-diarrheal treatment is ineffective)	Grade 1 or 2	Maintain dose; continue anti-diarrheal treatment. Loperamide (4 mg at first onset, followed by 2 mg every 2–4 hrs until diarrhea free for 12 hrs)
	Grade 3 or 4	Hold AZD6244 until < tolerable grade 2, loperamide; reduce 1 dose level and resume treatment [1].
	Recurrent Grade 3 or 4	Hold AZD6244 until < tolerable grade 2, loperamide; reduce 1 additional dose level and resume treatment [1].
	Recurrent Grade 3 or 4 after 2 dose reductions	Remove patient from study arm.
Hematologic (neutrophils, platelets, hemoglobin)	Grade 1 or 2	Maintain dose
	Grade 3 or 4	Hold AZD6244 until < grade 2, then reduce 1 dose level and resume treatment.
	Recurrent Grade 3 or 4	Hold AZD6244 until < grade 2, then reduce 1 additional dose level and resume treatment.
	Recurrent Grade 3 or 4 after 2 dose reductions	Remove patient from study arm.
Liver Function (serum bilirubin, AST, or ALT)	Grade 1 or 2	Maintain dose
	Grade 3 or 4	Hold AZD6244 until < grade 2, then reduce 1 dose level and resume treatment
	Recurrent Grade 3 or 4	Hold AZD6244 until < grade 2, then reduce 1 additional dose level and resume treatment
	Recurrent Grade 3 or 4 after 2 dose reductions	Remove patient from study arm.
Other non-	Grade 1 or 2	Maintain dose

hematological toxicity	Any Grade 2 of concern (e.g., prolonged cardiac, pulmonary, or neurotoxicity, intolerable stomatitis)	Hold AZD6244 until < grade 1, then reduce 1 dose level and resume treatment
	Grade 3 or 4	Hold AZD6244 until < tolerable grade 2, then reduce 1 dose level and resume treatment [2]
	Recurrent Grade 3 or 4	Hold AZD6244 until < tolerable grade 2, then reduce 1 additional dose level and resume treatment [2]
1 If event has not improved to < tolerable grade 2 or baseline within < 21 days, patient should be removed from the study. 2 Patients with medically concerning grade 3 or 4 AEs related to AZD 6244 may be taken off study at investigator's discretion.		

6.1.2 MK2206

Table 10: MK2206 Monotherapy Dose Level Reductions

Dose Level	**MK2206 Dose**
Starting Dose	200 mg QW
First Dose Reduction	135 mg QW
Second Dose Reduction	90 mg QW

Table 11: Dosage Modification Criteria for MK2206–related Toxicities

Event	**AE Grade or Observation**	**Dose modification**
Dermatology/Skin	Grade 1 or 2	Maintain dose
	Grade 3 or 4 or intolerable grade 2	Hold MK2206 until < tolerable grade 2, then reduce 1 dose level and resume treatment [1].
	Recurrent Grade 3	Hold MK2206 until < tolerable grade 2, then reduce 1 additional dose level and resume treatment [1].
	Recurrent Grade 3 after 2 dose reductions	Remove patient from study arm.
	Grade 4	Patients with grade 4 rash (i.e. life threatening generalized exfoliative ulceration; erythemia

		multiforme/toxic epidermal necrolysis should be removed from study arm.
Diarrhea (if anti-diarrheal treatment is ineffective)	Grade 1 or 2	Maintain dose; continue anti-diarrheal treatment. Loperamide (4 mg at first onset, followed by 2 mg every 2–4 hrs until diarrhea free for 12 hrs)
	Grade 3 or 4	Hold MK2206 until < tolerable grade 2, loperamide; reduce 1 dose level and resume treatment [1].
	Recurrent Grade 3 or 4	Hold MK2206 until < tolerable grade 2, loperamide; reduce 1 additional dose level and resume treatment [1].
	Recurrent Grade 3 or 4 after 2 dose reductions	Remove patient from study arm.
Hematologic (neutrophils, platelets, hemoglobin)	Grade 1 or 2	Maintain dose
	Grade 3 or 4	Hold MK2206 until < grade 2, then reduce 1 dose level and resume treatment.
	Recurrent Grade 3 or 4	Hold MK2206 until < grade 2, then reduce 1 additional dose level and resume treatment.
	Recurrent Grade 3 or 4 after 2 dose reductions	Remove patient from study arm.
Liver Function (serum bilirubin, AST, or ALT)	Grade 1 or 2	Maintain dose
	Grade 3 or 4	Hold MK2206 until < grade 2, then reduce 1 dose level and resume treatment
	Recurrent Grade 3 or 4	Hold MK2206 until < grade 2, then reduce 1 additional dose level and resume treatment
	Recurrent Grade 3 or 4 after 2 dose reductions	Remove patient from study arm.
Other non-hematological toxicity	Grade 1 or 2	Maintain dose
	Any Grade 2 of concern (e.g., prolonged cardiac, pulmonary, or neurotoxicity, intolerable stomatitis)	Hold MK2206 until < grade 1, then reduce 1 dose level and resume treatment
	Grade 3 or 4	Hold MK2206 until < tolerable grade 2, then reduce 1 dose level and resume treatment [2]
	Recurrent	Hold MK2206 until < tolerable grade 2, then reduce 1 additional dose level and resume

	Grade 3 or 4	treatment [2]
1 If event has not improved to < tolerable grade 2 or baseline within < 21 days, patient should be removed from the study. 2 Patients with medically concerning grade 3 or 4 AEs related to MK2206 may be taken off study at investigator's discretion.		

In the event of Grade 3 hyperglycemic events (>250 mg/dL) consult with an endocrinologist or other specialist. If glucose levels do not return to grade 1 or lower within one week of appropriate therapy, patients should be considered to have a DLT (dose modification, etc.). Appropriate therapy will usually involve oral antihyperglycemic agents, since the inhibition of glucose transport into the cell by AKT/mTOR inhibitors may render insulin ineffective. The goal of therapy is to keep fasting glucose <150 mg/dL, random blood glucose levels <180 mg/dL, and Hemoglobin A1c <8%. Glucose monitoring should be performed weekly, during the first cycle of therapy, and on day 1 of subsequent cycles, prior to drug administration. Hemoglobin A1c monitoring, for patients requiring treatment of hyperglycemia, should be performed with each cycle of MK-2206 therapy.

6.1.3 Lapatinib

Table 12: Lapatinib Monotherapy Dose Level Reductions

Dose Level	Lapatinib **Dose**
Starting Dose	1500 mg daily
First Dose Reduction	1250 mg daily
Second Dose Reduction	1000 mg daily

Table 13: Dosage Modification Criteria for Lapatinib –related Toxicities

Event	**AE Grade or Observation**	**Dose modification**
Dermatology/Skin	Grade 1 or 2	Maintain dose
	Grade 3 or 4 or intolerable grade 2	Hold Lapatinib until < tolerable grade 2, then reduce 1 dose level and resume treatment [1].
	Recurrent Grade 3	Hold Lapatinib until < tolerable grade 2, then reduce 1 additional dose level and resume treatment [1].
	Recurrent Grade 3 after 2 dose reductions	Remove patient from study arm.
	Grade 4	Patients with grade 4 rash (i.e. life threatening generalized exfoliative ulceration; erythemia multiforme/toxic epidermal necrolysis should

		be removed from study arm.
Diarrhea (if anti-diarrheal treatment is ineffective)	Grade 1 or 2	Maintain dose; continue anti-diarrheal treatment. Loperamide (4 mg at first onset, followed by 2 mg every 2–4 hrs until diarrhea free for 12 hrs)
	Grade 3 or 4	Hold Lapatinib until < tolerable grade 2, loperamide; reduce 1 dose level and resume treatment [1].
	Recurrent Grade 3 or 4	Hold Lapatinib until < tolerable grade 2, loperamide; reduce 1 additional dose level and resume treatment [1].
	Recurrent Grade 3 or 4 after 2 dose reductions	Remove patient from study arm.
Hematologic (neutrophils, platelets, hemoglobin)	Grade 1 or 2	Maintain dose
	Grade 3 or 4	Hold Lapatinib until < grade 2, then reduce 1 dose level and resume treatment.
	Recurrent Grade 3 or 4	Hold Lapatinib until < grade 2, then reduce 1 additional dose level and resume treatment.
	Recurrent Grade 3 or 4 after 2 dose reductions	Remove patient from study arm.
Liver Function (serum bilirubin, AST, or ALT)	Grade 1 or 2	Maintain dose
	Grade 3 or 4	Hold Lapatinib until < grade 2, then reduce 1 dose level and resume treatment
	Recurrent Grade 3 or 4	Hold Lapatinib until < grade 2, then reduce 1 additional dose level and resume treatment
	Recurrent Grade 3 or 4 after 2 dose reductions	Remove patient from study arm.
Other non-hematological toxicity	Grade 1 or 2	Maintain dose
	Any Grade 2 of concern (e.g., prolonged cardiac, pulmonary, or neurotoxicity, intolerable stomatitis)	Hold Lapatinib until < grade 1, then reduce 1 dose level and resume treatment
	Grade 3 or 4	Hold Lapatinib until < tolerable grade 2, then reduce 1 dose level and resume treatment [2]
	Recurrent Grade 3 or 4	Hold Lapatinib until < tolerable grade 2, then reduce 1 additional dose level and resume

		treatment [2]

1 If event has not improved to < tolerable grade 2 or baseline within < 21 days, patient should be removed from the study.

2 Patients with medically concerning grade 3 or 4 AEs related to Lapatinib may be taken off study at investigator's discretion.

6.1.4 Criteria for Evaluating Cardiac and Respiratory Events

A patient who has a ≥20% decrease in the left ventricular cardiac ejection fraction (as a % of baseline) that is asymptomatic, and the ejection fraction is below the institution's lower limit of normal, should have a repeat evaluation of ejection fraction 1-2 weeks later while still receiving therapy with lapatinib. If the repeat ejection fraction evaluation confirms a ≥ 20% decrease in left ventricular cardiac ejection fraction (as a % of baseline), and the ejection fraction is below the institution's lower limit of normal, then the CTEP medical monitor should be consulted to help determine if therapy should be temporarily discontinued. If therapy is interrupted and the left ventricular ejection fraction recovers during the next 3 weeks, in consultation with the CTEP medical monitor, the patient may be restarted on therapy with lapatinib at a reduced dose. For such patients, monitoring of left ventricular ejection fraction will then be performed 2 weeks and 4 weeks after re-challenge and then every 4 weeks therafter for at least 16 weeks or until resolution. If repeat ejection fraction evaluation still shows a decrease ≥20% in left ventricular ejection fraction (as a % of baseline), and the value is below the insitution's lower limit of normal, then the patient should be withdrawn from therapy with lapatinib.

Patients with and NCI-CTCAE grade 3 (LVEF less than 40% and symptomatic) or grade 4 (LVEF less than 20%) left ventricular systolic dysfunction or interstitial pneumonitis must be withdrawn from lapatinib therapy.

6.2 Other agents

6.2.1 Sunitinib

Table 14: Sunitinib Monotherapy Dose Level Reductions

Dose Level	Sunitinib **Dose**
Starting Dose	50 mg daily
First Dose Reduction	37.5 mg daily
Second Dose Reduction	25 mg daily

Table 15. Management of Treatment-Emergent Hypertension

Recommended Hypertension Monitoring and Management (BP in mmHg)

Grade (CTCAE v4)	**Antihypertensive Therapy**	**Blood Pressure Monitoring**	**Sunitinib Dose Modification**
Persistent Grade 1 Pre-hypertension Systolic 120-139 Diastolic 80-90		Standard	No change
Persistent Grade 2-Moderate Systolic 140-159 Diastolic 90-99 Protocol-specific guidance supersedes any other management guidelines, including CTCAE v4	Step 1) Initiate LA DHP CCB treatment and if needed, after 24-48 hr Rx, increase dose in stepwise fashion every 24-48 hours until BP is controlled or at max dose of Rx Step 2) If BP still not controlled, add another antihypertensive Rx, a BB, ACE1, ARB, or ABB; increase dose of this drug as described in step 1 Step 3) If BP still not controlled, add 3rd drug from the list of antihypertensives in step 2; increase dose of this drug as described in step 1 Step 4) If BP still not controlled, consider either 1 dose reduction of sunitinib or stopping sunitinib NOTE: Stopping or reducing the dose of sunitinib is expected to cause a decrease in BP. The treating physician should monitor the patient for hypotension and adjust the number and dose of antihypertensive medication(s) accordingly.	BP should be monitored as recommended by the treating physician	No change except as described in step 4

Persistent Grade 3 Severe Systolic >160 Diastolic >100 Protocol-specific guidance supersedes any other management guidelines, including CTCAE v4	HOLD sunitinib until systolic BP <159 and diastolic BP <99. BP management is identical to that for Grade 2 (see steps 1-4 above) with 2 major exceptions: 1) If systolic BP >180 or diastolic BP >110 and the patient is symptomatic: optimal management with intensive IV support in ICU; STOP sunitinib and notify hospital staff that stopping sunitinib may result in a decrease in BP and 2) If systolic BP >180 or diastolic BP >110 and the patient is asymptomatic, 2 new anti-hypertensives must be given together in step 1 (and dose escalated appropriately as in step 1). NOTE: Stopping or reducing the dose of sunitinib is expected to cause a decrease in BP. The treating physician should monitor the patient for hypotension and adjust the number and dose of antihypertensive medication(s) accordingly	BP should be monitored as recommended by the treating physician unless the patient is symptomatic with systolic BP >180 or diastolic BP >110 in which case, monitoring should be intensive.	HOLD sunitinib until systolic BP <159 and diastolic BP <99. In most circumstances, if BP cannot be controlled after an optimal trial of anti-hypertensive medications, consider either 1 dose reduction of sunitinib or stopping sunitinib. HOWEVER, if the patient requires hospitalization for management of symptomatic systolic BP >180 or diastolic BP >110,permanently discontinue sunitinib or if BP is controlled, re-start sunitinib at 1 lower dose level after consultation with the study Principal Investigator
Grade 4 Life-threatening consequences of hypertension	Optimal management with intensive IV support in ICU; STOP sunitinib and notify hospital staff that stopping sunitinib may result in a decrease in BP	Intensive	Permanently discontinue sunitinib or if BP is controlled, re-start sunitinib at 1 lower dose level after consultation with the study Principal Investigator
Abbreviations: dihydropyridine calcium-channel blockers (DHP-CCB), selective beta blockers (BB), angiotensin converting enzyme inhibitors (ACEI), angiotensin II receptor blockers (ARB), alpha beta blocker (ABB)			

If patients require a delay of >2 weeks for management of hypertension, discontinue protocol therapy

If patients require >2 dose reductions, discontinue protocol therapy

Patients may have up to 2 drugs for management of hypertension prior to any dose reduction in sunitinib

24-48 hours should elapse between modifications of anti-hypertensive therapy

Hypertension should be graded using CTCAE v4.

Increases in blood pressure (BP) and cases of hypertension have been associated with many drugs acting on the VEGF pathway. The proposed mechanism for this increase is through inhibition of VEGF-induced peripheral vasodilation. Hypertension following sunitinib treatment has rarely been seen in animal studies or clinical trials. Specific guidelines for management of this adverse event and a table of various antihypertensive medications are provided in **Appendix H**. In addition, guidance on the collection and recording of BP information is also provided.

Table 16. Dosage Modification Criteria for Sunitinib –related Toxicities

Event	**AE Grade or Observation**	**Dose modification**
Neutropenia	Grades 1 and 2	Maintain dose
	Grade 3*	Hold sunitinib until < grade 2, then resume at same dose level
	Grade 4	Hold sunitinib until < grade 2, then reduce 1 dose level and resume treatment
Thrombocytopenia	Grades 1 and 2	Maintain dose
	Grade 3*	Hold sunitinib until < grade 2, then reduce 1 dose level and resume treatment
	Grade 4	Hold sunitinib until < grade 2, then reduce 1 dose level and resume treatment
Fever or flu-like symptoms	Grades 1-4	Maintain dose
Fatigue (lethargy, malaise, asthenia)	Grades 1 and 2	Maintain dose
	Grade 3*	Hold sunitinib until < grade 2, then reduce 1 dose level and resume treatment
	>450 but < 550 msec	Review patient's concomitant medications for QT interval-prolonging agents. Correct any electrolyte abnormalities. Continue sunitinib at current dose level.

QTc prolongation Do not use CTCAE v4 grades	> 550 msec	Stop sunitinib and any other QT-interval prolonging agents immediately. Correct any electrolyte abnormalities, then If there is a plausible explanation for AE other than sunitinib treatment, resume sunitinib at current dose level. If sunitinib may have contributed to the AE: Reduce 2 dose levels and restart sunitinib. If QTc remains <500 msec after 14 days at reduced dose, increase one dose level and continue sunitinib. If QTc remains <500 msec after 14 days, original dose of sunitinib may be resumed.
Hand-foot syndrome	Grades 1 and 2	Maintain dose
	Grade 3*	Hold sunitinib until < grade 1, then resume treatment at same dose or reduce 1 dose level
AST and/or ALT elevation (SGOT, SGPT)	Grades 1-4	Maintain dose *

*Recurrent grade 3 events require dose reduction.

Table 17. Management of Other Clinically Significant Sunitinib-related AEs (not specifically addressed above)

Observation	**Action**
AE resolves promptly with supportive care	Maintain dose level
1. Grade 3 or higher (non-hematologic or grade 4 (hematologic) AE related to sunitinib and lasting >5 days that does not resolve to grade 2 or below despite maximum supportive care for < 48 hours. 2. Lower grade but related AEs (e.g., creatinine)	Reduce one dose level
AE does not resolve to grade 2 or below after treating patient at the lowest (i.e., 25* mg or 12.5* daily) reduced dose level.	In general, remove patient from study**
* 25 mg if the patients start dosing at 50 mg on a 4/2 schedule (solid tumors) or 12.5 mg if patients start dosing at 37.5 mg ** After consultation with study sponsor (DCTD, NCI), a dose of 25 mg daily may be	

considered for patients on study > 3 months who are benefiting from the agent.

6.3 Treatment delays and modifications for administrative needs

Brief interruptions and delays may occasionally be required due to travel delays, airport closure, inclement weather, family responsibilities, security alerts, and government holidays, etc. These delays will not be considered protocol deviations, and will not be separately reported. A patient that interrupts therapy for more than 21 days will be taken off treatment.

6.4 Treatment delays and modifications for medical needs

Patients experiencing complications of their disease or other medical illness not attributable to disease progression, or protocol therapy may also require brief interruptions and delays that will not be considered protocol deviations, and will not be separately reported. A patient that interrupts therapy for more than 21 days will be taken off treatment.

7 ADVERSE EVENTS: LIST AND REPORTING REQUIREMENTS

Adverse event monitoring and reporting is a routine part of every clinical trial. The following list of adverse events (Section 7.3) and the characteristics of an observed adverse event (Section 7.4) will determine whether the event requires expedited (via CTEP-AERS; Section 7.5) or routine (via CTMS or CDUS; Section 7.6) reporting.

The following sections related to adverse event monitoring and reporting apply only to patients receiving treatment under one of the experimental arms (Erlotinib, AZD6244, MK2206, Lapatinib and Sunitinib). No toxicity data will be reported to CTEP for patients being followed under the NOS arm or individuals enrolled for germline mutation testing and follow-up only; applicable side effects occurring in these participants will be reported to the IRB.

7.1 Comprehensive Adverse Events and Potential Risks Lists (CAEPRs)

The Comprehensive Adverse Event and Potential Risks list (CAEPR) was developed to provide a single, complete list of reported and/or potential adverse events associated with an agent using a uniform presentation of adverse events by body system. In addition to the comprehensive list, the subset of those events that are "expected" [i.e., the Agent Specific Adverse Event List (ASAEL)] is presented in a separate column and identified with bold and italicized text. This subset is used to guide expedited reporting requirements.

7.1.1 CAEPRs for CTEP-Supplied Investigational Agent(s)

7.1.1.1 Erlotinib (OSI-774, NSC 718781)

Comprehensive Adverse Events and Potential Risks list (CAEPR)

for

Erlotinib (Tarceva®, NSC 718781)

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 3622 patients.* Below is the CAEPR for erlotinib (Tarceva®).

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 2.5, March 25, 2015[77]

Adverse Events with Possible Relationship to Erlotinib (Tarceva®) (CTCAE 4.0 Term) [n= 3622]			Specific Protocol Exceptions to Expedited Reporting (SPEER)
Likely (>20%)	**Less Likely (<=20%)**	**Rare but Serious (<3%)**	
BLOOD AND LYMPHATIC SYSTEM DISORDERS			
		Disseminated intravascular coagulation[104]	
		Hemolytic uremic syndrome[104]	
		Thrombotic thrombocytopenic purpura[104]	
CARDIAC DISORDERS			
		Myocardial infarction[104]	
EYE DISORDERS			
	Conjunctivitis		***Conjunctivitis (Gr 2)***
	Dry eye		***Dry eye (Gr 2)***
		Eye disorders - Other (corneal perforation)	
	Eye disorders - Other (eyelash in-growth and/or thickening)		
		Keratitis	
GASTROINTESTINAL DISORDERS			
	Abdominal pain		***Abdominal pain (Gr 3)***
Diarrhea			***Diarrhea (Gr 3)***
	Dry mouth		***Dry mouth (Gr 2)***
	Dyspepsia		***Dyspepsia (Gr 2)***
	Gastrointestinal hemorrhage[105]		
		Gastrointestinal perforation[106]	
	Mucositis oral		***Mucositis oral (Gr 3)***
	Nausea		***Nausea (Gr 3)***
Vomiting			***Vomiting (Gr 3)***
GENERAL DISORDERS AND ADMINISTRATION SITE CONDITIONS			
Fatigue			***Fatigue (Gr 3)***
HEPATOBILIARY DISORDERS			
		Hepatic failure	
INFECTIONS AND INFESTATIONS			
	Skin infection[107]		***Skin infection[107] (Gr 2)***
INVESTIGATIONS			

Adverse Events with Possible Relationship to Erlotinib (Tarceva®) (CTCAE 4.0 Term) [n= 3622]			Specific Protocol Exceptions to Expedited Reporting (SPEER)
Likely (>20%)	**Less Likely (<=20%)**	**Rare but Serious (<3%)**	
	Alanine aminotransferase increased		***Alanine aminotransferase increased (Gr 3)***
	Alkaline phosphatase increased		
	Aspartate aminotransferase increased		***Aspartate aminotransferase increased (Gr 3)***
	Blood bilirubin increased		***Blood bilirubin increased (Gr 3)***
METABOLISM AND NUTRITION DISORDERS			
Anorexia			***Anorexia (Gr 3)***
	Dehydration		***Dehydration (Gr 3)***
NERVOUS SYSTEM DISORDERS			
	Dysgeusia		***Dysgeusia (Gr 2)***
	Headache		***Headache (Gr 2)***
		Intracranial hemorrhage	
		Ischemia cerebrovascular[104]	
RENAL AND URINARY DISORDERS			
		Acute kidney injury	
RESPIRATORY, THORACIC AND MEDIASTINAL DISORDERS			
	Cough		***Cough (Gr 2)***
	Dyspnea		***Dyspnea (Gr 3)***
	Epistaxis		
	Pneumonitis		***Pneumonitis (Gr 3)***
SKIN AND SUBCUTANEOUS TISSUE DISORDERS			
	Alopecia		***Alopecia (Gr 2)***
	Dry skin		***Dry skin (Gr 2)***
		Erythema multiforme	
	Nail loss		***Nail loss (Gr 2)***
		Palmar-plantar erythrodysesthesia syndrome	
	Pruritus		***Pruritus (Gr 2)***
	Rash acneiform		***Rash acneiform (Gr 2)***
Rash maculo-papular			***Rash maculo-papular (Gr 3)***
		Stevens-Johnson syndrome	
		Toxic epidermal necrolysis	

[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]The risk of myocardial infarction, cerebrovascular accident, and microangiopathic hemolytic anemia is increased in patients with pancreatic cancer who were treated concomitantly with gemcitabine.

[3]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.

[4]Gastrointestinal perforation includes Colonic perforation, Duodenal perforation, Esophageal perforation, Gastric perforation, Ileal perforation, Jejunal perforation, Rectal perforation, and Small intestinal perforation under the GASTROINTESTINAL DISORDERS SOC.

[5]Includes infection of the skin (folliculitis or cellulitis) as complications of rash.

Adverse events reported on erlotinib (Tarceva®) trials, but for which there is insufficient evidence to suggest that there was a reasonable possibility that erlotinib (Tarceva®) caused the adverse event:

EYE DISORDERS - Blurred vision; Eye disorders - Other (orbital cellulitis); Uveitis; Watering eyes
GASTROINTESTINAL DISORDERS - Colitis; Constipation; Duodenal ulcer; Dysphagia; Esophagitis; Gastric ulcer; Gastritis; Gastrointestinal disorders - Other (pneumatosis intestinalis); Pancreatitis
GENERAL DISORDERS AND ADMINISTRATION SITE CONDITIONS - Edema limbs
HEPATOBILIARY DISORDERS - Cholecystitis
INVESTIGATIONS - Creatinine increased; INR increased (in patients taking Coumadin); Lymphocyte count decreased; Platelet count decreased
METABOLISM AND NUTRITION DISORDERS - Hyperglycemia; Hyperkalemia; Hypocalcemia; Hypokalemia; Hypomagnesemia; Hyponatremia; Hypophosphatemia
MUSCULOSKELETAL AND CONNECTIVE TISSUE DISORDERS - Generalized muscle weakness
NERVOUS SYSTEM DISORDERS - Dizziness; Peripheral sensory neuropathy
PSYCHIATRIC DISORDERS - Confusion
RESPIRATORY, THORACIC AND MEDIASTINAL DISORDERS - Adult respiratory distress syndrome; Pharyngolaryngeal pain
SKIN AND SUBCUTANEOUS TISSUE DISORDERS - Urticaria
VASCULAR DISORDERS - Thromboembolic event

Note: Erlotinib (Tarceva®) 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.

Note: Erlotinib (Tarceva®)-induced diarrhea and/or vomiting has been associated with dehydration, hyperkalemia; hypocalcemia; hypokalemia; hypomagnesemia; hyponatremia; hypophosphatemia, increased creatinine, and renal failure.

Note: Cases of hepatic failure and hepatorenal syndrome (including fatalities) have been reported during use of erlotinib (Tarceva®) in patients with or without baseline hepatic impairment.

7.1.1.2 Comprehensive Adverse Events and Potential Risks list (CAEPR) for Selumetinib (AZD6244 Free base [NSC 741078], AZD6244 Hydrogen sulfate [NSC 748727])

The Comprehensive Adverse Event 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 via CTEP-AERS (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 665 patients.* Below is the CAEPR for Selumetinib (AZD6244).

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 2.4, February 26, 2013[1]

Adverse Events with Possible Relationship to Selumetinib (AZD6244) (CTCAE 4.0 Term) [n= 665]			**Specific Protocol Exceptions to Expedited Reporting (SPEER)** (formerly known as ASAEL)
Likely (>20%)	**Less Likely (<=20%)**	**Rare but Serious (<3%)**	
BLOOD AND LYMPHATIC SYSTEM DISORDERS			
	Anemia		***Anemia (Gr 3)***
		Febrile neutropenia[2]	
CARDIAC DISORDERS			
		Left ventricular systolic dysfunction	***Left ventricular systolic dysfunction (Gr 2)***
GASTROINTESTINAL DISORDERS			
	Abdominal pain		***Abdominal pain (Gr 2)***
	Constipation		***Constipation (Gr 2)***
Diarrhea[3]			***Diarrhea[3] (Gr 3)***
	Dry mouth		
	Mucositis oral		***Mucositis oral (Gr 2)***
Nausea			***Nausea (Gr 3)***
Vomiting			***Vomiting (Gr 2)***
GENERAL DISORDERS AND ADMINISTRATION SITE CONDITIONS			
	Edema face		***Edema face (Gr 2)***
Edema limbs			***Edema limbs (Gr 2)***
Fatigue			***Fatigue (Gr 2)***
	Fever		***Fever (Gr 2)***
INVESTIGATIONS			
	Alanine aminotransferase increased		***Alanine aminotransferase increased (Gr 3)***
	Alkaline phosphatase increased		
	Aspartate aminotransferase increased		***Aspartate aminotransferase increased (Gr 3)***
	Platelet count decreased		
METABOLISM AND NUTRITION DISORDERS			
	Anorexia		***Anorexia (Gr 2)***
	Hypoalbuminemia		
	Hypomagnesemia		
NERVOUS SYSTEM DISORDERS			
	Dizziness		
	Headache		***Headache (Gr 2)***
PSYCHIATRIC DISORDERS			
	Insomnia		
RESPIRATORY, THORACIC AND MEDIASTINAL DISORDERS			

	Cough		***Cough (Gr 2)***
	Dyspnea		***Dyspnea (Gr 2)***
SKIN AND SUBCUTANEOUS TISSUE DISORDERS			
	Dry skin		***Dry skin (Gr 2)***
	Pruritus		
Rash acneiform			***Rash acneiform (Gr 3)***
Rash maculo-papular			***Rash maculo-papular (Gr 2)***
VASCULAR DISORDERS			
	Hypertension		

[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]Febrile neutropenia/neutropenic infection has been observed primarily in trials combining Selumetinib (AZD6244) and docetaxel.

[3]SBE-CD (Captisol®, vehicle) in the mix and drink formulation is known to cause soft stools and/or diarrhea in rats and dogs; however, it is possible that some of these findings might be related to exacerbation of the vehicle effect by Selumetinib (AZD6244).

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

Also reported on Selumetinib (AZD6244) trials but with the relationship to Selumetinib (AZD6244) still undetermined:

BLOOD AND LYMPHATIC SYSTEM DISORDERS - Blood and lymphatic system disorders - Other (hemorrhagic anemia)

CARDIAC DISORDERS - Acute coronary syndrome; Cardiac disorders - Other (Takatsubo cardiomyopathy syndrome); Chest pain - cardiac; Heart failure; Palpitations; Right ventricular dysfunction; Sinus bradycardia

EYE DISORDERS - Blurred vision; Extraocular muscle paresis; Eye disorders - Other (bilateral macular edema); Eye disorders - Other (black haze in line of vision); Eye disorders - Other (elevated intraocular pressure); Flashing lights; Glaucoma; Retinopathy

GASTROINTESTINAL DISORDERS - Abdominal distension; Ascites; Colitis; Dyspepsia; Esophagitis; Flatulence; Gastric hemorrhage; Gastroesophageal reflux disease; Ileal stenosis

GENERAL DISORDERS AND ADMINISTRATION SITE CONDITIONS - Flu like symptoms; Non-cardiac chest pain

HEPATOBILIARY DISORDERS - Hepatic failure; Hepatobiliary disorders - Other (cholangitis)

INFECTIONS AND INFESTATIONS - Infection[4]

INVESTIGATIONS - CPK increased; Creatinine increased; Electrocardiogram QT corrected interval prolonged; Neutrophil count decreased; Weight gain

METABOLISM AND NUTRITION DISORDERS - Dehydration; Hypokalemia; Hyponatremia

MUSCULOSKELETAL AND CONNECTIVE TISSUE DISORDERS - Arthralgia; Back pain; Generalized muscle weakness; Musculoskeletal and connective tissue disorder - Other (joint swelling); Musculoskeletal and connective tissue disorder - Other (rhabdomyolysis); Myalgia; Myositis; Pain in extremity

NERVOUS SYSTEM DISORDERS - Cognitive disturbance; Depressed level of consciousness; Dysgeusia; Dysphasia; Ischemia cerebrovascular; Leukoencephalopathy; Memory impairment; Nervous system disorders -

Other (numbness); Nervous system disorders - Other (spinal cord compression); Oculomotor nerve disorder; Peripheral sensory neuropathy; Reversible posterior leukoencephalopathy syndrome; Seizure

PSYCHIATRIC DISORDERS - Confusion; Depression

RENAL AND URINARY DISORDERS - Acute kidney injury; Proteinuria

RESPIRATORY, THORACIC AND MEDIASTINAL DISORDERS - Adult respiratory distress syndrome; Epistaxis; Hypoxia; Pharyngolaryngeal pain; Pleural effusion; Pneumonitis; Sore throat; Voice alteration

SKIN AND SUBCUTANEOUS TISSUE DISORDERS - Alopecia; Pain of skin; Palmar-plantar erythrodysesthesia syndrome; Photosensitivity; Scalp pain; Skin and subcutaneous tissue disorders - Other (angular cheilitis, unilateral); Skin and subcutaneous tissue disorders - Other (skin fissures)

VASCULAR DISORDERS - Hypotension; Thromboembolic event

Note: Selumetinib (AZD6244) 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.

7.1.1.3 Comprehensive Adverse Event and Potential Risks list (CAEPR) for MK-2206 (NSC 749607)

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 245 patients. Below is the CAEPR for MK-2206.

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 2.2, June 16, 2015[77]

Adverse Events with Possible Relationship to MK-2206 (CTCAE 4.0 Term) [n= 245]			Specific Protocol Exceptions to Expedited Reporting (SPEER)
Likely (>20%)	**Less Likely (<=20%)**	**Rare but Serious (<3%)**	
BLOOD AND LYMPHATIC SYSTEM DISORDERS			
	Anemia		
CARDIAC DISORDERS			
	Sinus bradycardia		***Sinus bradycardia (Gr 2)***
GASTROINTESTINAL DISORDERS			
	Diarrhea		***Diarrhea (Gr 3)***
	Mucositis oral		***Mucositis oral (Gr 3)***
Nausea			***Nausea (Gr 2)***
	Vomiting		***Vomiting (Gr 3)***

Adverse Events with Possible Relationship to MK-2206 (CTCAE 4.0 Term) [n= 245]			Specific Protocol Exceptions to Expedited Reporting (SPEER)
Likely (>20%)	**Less Likely (<=20%)**	**Rare but Serious (<3%)**	
GENERAL DISORDERS AND ADMINISTRATION SITE CONDITIONS			
Fatigue			***Fatigue (Gr 3)***
	Fever		***Fever (Gr 2)***
INVESTIGATIONS			
	Alanine aminotransferase increased		
	Alkaline phosphatase increased		
	Aspartate aminotransferase increased		
	Creatinine increased		
	Electrocardiogram QT corrected interval prolonged		***Electrocardiogram QT corrected interval prolonged (Gr 2)***
	Hemoglobin increased		
	Investigations - Other (eosinophilia)		***Investigations - Other (eosinophilia) (Gr 2)***
	Investigations - Other (insulin c-peptide increased)		
	Lymphocyte count decreased		***Lymphocyte count decreased (Gr 4)***
	Neutrophil count decreased		***Neutrophil count decreased (Gr 4)***
	Platelet count decreased		
	Weight loss		
	White blood cell decreased		***White blood cell decreased (Gr 2)***
METABOLISM AND NUTRITION DISORDERS			
	Anorexia		***Anorexia (Gr 2)***
Hyperglycemia			***Hyperglycemia (Gr 3)***
	Hypocalcemia		
	Hyponatremia		
NERVOUS SYSTEM DISORDERS			
	Dysgeusia		
	Headache		***Headache (Gr 2)***
SKIN AND SUBCUTANEOUS TISSUE DISORDERS			
	Dry skin		***Dry skin (Gr 2)***
	Pruritus		***Pruritus (Gr 2)***
Rash maculo-papular			***Rash maculo-papular (Gr 3)***

[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.

Adverse events reported on MK-2206 trials, but for which there is insufficient evidence to suggest that there was a reasonable possibility that MK-2206 caused the adverse event:

Blood and lymphatic system disorders - Febrile neutropenia; Hemolysis; Hemolytic uremic syndrome; Leukocytosis

Cardiac disorders - Atrioventricular block complete; Left ventricular systolic dysfunction; Myocardial infarction; Palpitations; Ventricular arrhythmia

Ear and labyrinth disorders - Vertigo

Endocrine disorders - Hypothyroidism

Eye disorders - Blurred vision; Conjunctivitis; Dry eye; Extraocular muscle paresis; Eye disorders - Other (blepharitis); Eye disorders - Other (eye swelling); Eye disorders - Other (foreign body sensation in eyes); Eye disorders - Other (iritis); Eye disorders - Other (mydriasis); Eye disorders - Other (visual acuity reduced); Eye pain; Floaters; Keratitis; Photophobia; Retinal detachment; Uveitis

Gastrointestinal disorders - Abdominal distension; Abdominal pain; Ascites; Cheilitis; Colonic perforation; Constipation; Dry mouth; Dyspepsia; Dysphagia; Gastritis; Gastrointestinal disorders - Other (oropharyngeal pain); Lip pain; Toothache

General disorders and administration site conditions - Chills; Edema face; Edema limbs; Flu like symptoms; General disorders and administration site conditions - Other (throat tightness); Injection site reaction; Irritability; Localized edema; Non-cardiac chest pain; Pain

Immune system disorders - Allergic reaction

Infections and infestations - Infections and infestations - Other (herpetic vesicular rash [due to herpes zoster infection]); Infections and infestations - Other (oral herpes); Lung infection; Nail infection; Paronychia; Pharyngitis; Rhinitis infective; Sepsis; Sinusitis; Skin infection; Urinary tract infection

Injury, poisoning and procedural complications - Fall

Investigations - Activated partial thromboplastin time prolonged; Blood bilirubin increased; INR increased; Investigations - Other (blood LDH increased); Investigations - Other (hyperinsulinemia); Lipase increased; Serum amylase increased; Weight gain

Metabolism and nutrition disorders - Dehydration; Hypercalcemia; Hyperkalemia; Hypermagnesemia; Hypoalbuminemia; Hypokalemia; Hypomagnesemia; Hypophosphatemia

Musculoskeletal and connective tissue disorders - Arthralgia; Back pain; Bone pain; Generalized muscle weakness; Myalgia; Neck pain; Pain in extremity

Nervous system disorders - Akathisia; Dizziness; Encephalopathy; Lethargy; Presyncope; Reversible posterior leukoencephalopathy syndrome; Seizure; Syncope; Tremor

Psychiatric disorders - Anxiety; Confusion; Depression; Insomnia

Renal and urinary disorders - Acute kidney injury; Proteinuria; Renal and urinary disorders - Other (renal tubular necrosis); Renal and urinary disorders - Other (glucose urine present); Urinary incontinence; Urinary tract pain

Reproductive system and breast disorders - Gynecomastia; Vaginal hemorrhage; Vaginal perforation

Respiratory, thoracic and mediastinal disorders - Bronchial obstruction; Cough; Dyspnea; Epistaxis; Hypoxia; Pneumonitis; Pulmonary edema; Respiratory failure; Sore throat

Skin and subcutaneous tissue disorders - Alopecia; Erythema multiforme; Erythroderma; Hyperhidrosis; Palmar-plantar erythrodysesthesia syndrome; Photosensitivity; Purpura; Rash acneiform; Skin and subcutaneous tissue disorders - Other (skin irritation); Stevens-Johnson syndrome; Urticaria

Vascular disorders - Hematoma; Hypertension; Hypotension; Thromboembolic event

Note: MK-2206 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.

7.1.1.4 Comprehensive Adverse Event and Potential Risks list (CAEPR) for GW572016 (Lapatinib, Tykerb, NSC 727989)

The Comprehensive Adverse Event 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 via CTEP-AERS (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 1890 patients.* Below is the CAEPR for GW572016 (lapatinib, Tykerb).

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, February 28, 2013[1]

Adverse Events with Possible Relationship to GW572016 (lapatinib, Tykerb) (CTCAE 4.0 Term) [n= 1890]			**Specific Protocol Exceptions to Expedited Reporting (SPEER)**
Likely (>20%)	**Less Likely (<=20%)**	**Rare but Serious (<3%)**	
CARDIAC DISORDERS			
		Left ventricular systolic dysfunction	***Left ventricular systolic dysfunction (Gr 2)***
GASTROINTESTINAL DISORDERS			
	Abdominal distension		***Abdominal distension (Gr 2)***
	Abdominal pain		***Abdominal pain (Gr 2)***
	Anal mucositis		***Anal mucositis (Gr 2)***
Diarrhea			***Diarrhea (Gr 3)***
	Dyspepsia		***Dyspepsia (Gr 2)***
	Flatulence		***Flatulence (Gr 2)***
	Mucositis oral		***Mucositis oral (Gr 2)***
Nausea			***Nausea (Gr 3)***
	Rectal mucositis		***Rectal mucositis (Gr 2)***
	Small intestinal mucositis		***Small intestinal mucositis (Gr 2)***
	Vomiting		***Vomiting (Gr 2)***
GENERAL DISORDERS AND ADMINISTRATION SITE CONDITIONS			
	Fatigue		***Fatigue (Gr 2)***
	Flu like symptoms		***Flu like symptoms (Gr 2)***
HEPATOBILIARY DISORDERS			
		Hepatic failure	***Hepatic failure (Gr 2)***
IMMUNE SYSTEM DISORDERS			
		Allergic reaction	

INVESTIGATIONS			
	Alanine aminotransferase increased		***Alanine aminotransferase increased (Gr 2)***
	Aspartate aminotransferase increased		***Aspartate aminotransferase increased (Gr 2)***
	Blood bilirubin increased		***Blood bilirubin increased (Gr 2)***
		Electrocardiogram QT corrected interval prolonged	***Electrocardiogram QT corrected interval prolonged (Gr 2)***
METABOLISM AND NUTRITION DISORDERS			
	Anorexia		***Anorexia (Gr 2)***
	Dehydration		***Dehydration (Gr 2)***
NERVOUS SYSTEM DISORDERS			
	Dysgeusia		***Dysgeusia (Gr 2)***
	Headache		***Headache (Gr 2)***
RESPIRATORY, THORACIC AND MEDIASTINAL DISORDERS			
	Laryngeal mucositis		***Laryngeal mucositis (Gr 2)***
	Pharyngeal mucositis		***Pharyngeal mucositis (Gr 2)***
		Pneumonitis	
	Tracheal mucositis		***Tracheal mucositis (Gr 2)***
SKIN AND SUBCUTANEOUS TISSUE DISORDERS			
	Nail loss		
	Pruritus		***Pruritus (Gr 2)***
	Rash acneiform		***Rash acneiform (Gr 2)***
Rash maculo-papular			***Rash maculo-papular (Gr 2)***
VASCULAR DISORDERS			
	Flushing		***Flushing (Gr 2)***

[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]Gastrointestinal obstruction includes Colonic obstruction, Duodenal obstruction, Esophageal obstruction, Ileal obstruction, Jejunal obstruction, Obstruction gastric, Rectal obstruction, and Small intestinal obstruction under the GASTROINTESTINAL DISORDERS SOC.

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

Also reported on GW572016 (lapatinib, Tykerb) trials but with the relationship to GW572016 (lapatinib, Tykerb) still undetermined:

BLOOD AND LYMPHATIC SYSTEM DISORDERS - Anemia; Febrile neutropenia

CARDIAC DISORDERS - Atrial fibrillation; Restrictive cardiomyopathy

EYE DISORDERS - Blurred vision

GASTROINTESTINAL DISORDERS - Constipation; Dysphagia; Gastritis; Gastrointestinal hemorrhage[2]; Gastrointestinal obstruction[3]

GENERAL DISORDERS AND ADMINISTRATION SITE CONDITIONS - Edema limbs; Fever; Pain

INFECTIONS AND INFESTATIONS – Infection[4]

INVESTIGATIONS - Alkaline phosphatase increased; Creatinine increased; Lymphocyte count decreased; Neutrophil count decreased; Platelet count decreased; Weight loss; White blood cell decreased

METABOLISM AND NUTRITION DISORDERS - Hyperglycemia; Hypoalbuminemia; Hypoglycemia; Hypokalemia; Hyponatremia; Hypophosphatemia

MUSCULOSKELETAL AND CONNECTIVE TISSUE DISORDERS - Arthralgia; Back pain; Myalgia

NERVOUS SYSTEM DISORDERS - Cerebrospinal fluid leakage; Depressed level of consciousness; Dizziness; Intracranial hemorrhage

PSYCHIATRIC DISORDERS - Insomnia

RENAL AND URINARY DISORDERS - Acute kidney injury

RESPIRATORY, THORACIC AND MEDIASTINAL DISORDERS - Cough; Dyspnea; Epistaxis; Pleural effusion

SKIN AND SUBCUTANEOUS TISSUE DISORDERS - Alopecia; Dry skin; Palmar-plantar erythrodysesthesia syndrome; Skin and subcutaneous tissue disorders - Other (seborrheic dermatitis); Urticaria

VASCULAR DISORDERS - Hypotension; Thromboembolic event; Vascular disorders - Other (hypovolemia)

Note: GW572016 (lapatinib, Tykerb) 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.

7.1.2 Adverse Event List(s) for Commercial Agent(s)

7.1.2.1 Sunitinib Malate (SU011248 L-malate, NSC 736511)

Comprehensive Adverse Events and Potential Risks list (CAEPR)

for

Sunitinib malate (SU011248 L-malate, NSC 736511)

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 7115 patients.* Below is the CAEPR for Sunitinib malate (SU011248 L-malate).

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 2.12, January 14, 2016(108)

Adverse Events with Possible Relationship to Sunitinib malate (SU011248 L-malate) (CTCAE 4.0 Term) [n= 7115]			**Specific Protocol Exceptions to Expedited Reporting (SPEER)**
Likely (>20%)	**Less Likely (<=20%)**	**Rare but Serious (<3%)**	
BLOOD AND LYMPHATIC SYSTEM DISORDERS			
	Anemia		***Anemia (Gr 3)***
		Hemolytic uremic syndrome	
		Thrombotic thrombocytopenic purpura	
CARDIAC DISORDERS			
		Cardiac disorders - Other (cardiomyopathy)	
		Heart failure	
		Left ventricular systolic dysfunction	
		Myocardial infarction	
ENDOCRINE DISORDERS			
		Endocrine disorders - Other (thyroiditis)	
		Hyperthyroidism	
	Hypothyroidism		***Hypothyroidism (Gr 2)***
EYE DISORDERS			
		Eye disorders - Other (macular edema)	***Eye disorders - Other (macular edema) (Gr 2)***
		Eye disorders - Other (vision deterioration)	***Eye disorders - Other (vision deterioration) (Gr 2)***
	Papilledema		***Papilledema (Gr 2)***
GASTROINTESTINAL DISORDERS			
	Abdominal distension		***Abdominal distension (Gr 2)***
Abdominal pain			***Abdominal pain (Gr 3)***
Anal mucositis			***Anal mucositis (Gr 2)***
Constipation			***Constipation (Gr 2)***
Diarrhea			***Diarrhea (Gr 3)***
	Dry mouth		***Dry mouth (Gr 2)***
Dyspepsia			***Dyspepsia (Gr 2)***
		Esophagitis	
	Flatulence		***Flatulence (Gr 2)***
	Gastritis		***Gastritis (Gr 2)***
	Gastroesophageal reflux disease		
		Gastrointestinal perforation(109)	
Mucositis oral			***Mucositis oral (Gr 3)***

Adverse Events with Possible Relationship to Sunitinib malate (SU011248 L-malate) (CTCAE 4.0 Term) [n= 7115]			**Specific Protocol Exceptions to Expedited Reporting (SPEER)**
Likely (>20%)	**Less Likely (<=20%)**	**Rare but Serious (<3%)**	
Nausea			***Nausea (Gr 3)***
	Oral pain		***Oral pain (Gr 2)***
		Pancreatitis	
Rectal mucositis			***Rectal mucositis (Gr 2)***
Small intestinal mucositis			***Small intestinal mucositis (Gr 2)***
Vomiting			***Vomiting (Gr 3)***
GENERAL DISORDERS AND ADMINISTRATION SITE CONDITIONS			
	Chills		***Chills (Gr 2)***
	Edema limbs		***Edema limbs (Gr 2)***
Fatigue			***Fatigue (Gr 3)***
	Fever		***Fever (Gr 2)***
	Non-cardiac chest pain		***Non-cardiac chest pain (Gr 2)***
HEPATOBILIARY DISORDERS			
		Cholecystitis	
		Hepatic failure	
IMMUNE SYSTEM DISORDERS			
		Allergic reaction(110)	
INFECTIONS AND INFESTATIONS			
		Infection and infestations - Other (necrotizing fasciitis)	
INJURY, POISONING AND PROCEDURAL COMPLICATIONS			
		Wound complication	
INVESTIGATIONS			
	Alanine aminotransferase increased		***Alanine aminotransferase increased (Gr 3)***
	Alkaline phosphatase increased		***Alkaline phosphatase increased (Gr 2)***
	Aspartate aminotransferase increased		***Aspartate aminotransferase increased (Gr 3)***
	Blood bilirubin increased		***Blood bilirubin increased (Gr 2)***
	CPK increased		
	Creatinine increased		***Creatinine increased (Gr 3)***
		Electrocardiogram QT corrected interval prolonged	
	Lipase increased		***Lipase increased (Gr 4)***
	Lymphocyte count decreased		***Lymphocyte count decreased (Gr 2)***
	Neutrophil count decreased		***Neutrophil count decreased (Gr 4)***
	Platelet count decreased		***Platelet count decreased (Gr 4)***

Adverse Events with Possible Relationship to Sunitinib malate (SU011248 L-malate) (CTCAE 4.0 Term) [n= 7115]			Specific Protocol Exceptions to Expedited Reporting (SPEER)
Likely (>20%)	**Less Likely (<=20%)**	**Rare but Serious (<3%)**	
	Serum amylase increased		***Serum amylase increased (Gr 2)***
	Weight loss		***Weight loss (Gr 2)***
	White blood cell decreased		***White blood cell decreased (Gr 3)***
METABOLISM AND NUTRITION DISORDERS			
Anorexia			***Anorexia (Gr 3)***
	Dehydration		***Dehydration (Gr 3)***
	Hyperuricemia		***Hyperuricemia (Gr 2)***
	Hypoalbuminemia		***Hypoalbuminemia (Gr 2)***
		Hypoglycemia	
	Hypophosphatemia		***Hypophosphatemia (Gr 2)***
		Tumor lysis syndrome	
MUSCULOSKELETAL AND CONNECTIVE TISSUE DISORDERS			
	Arthralgia		***Arthralgia (Gr 2)***
	Back pain		***Back pain (Gr 2)***
		Musculoskeletal and connective tissue disorder - Other (rhabdomyolysis)	
	Myalgia		***Myalgia (Gr 2)***
		Osteonecrosis of jaw	
	Pain in extremity		***Pain in extremity (Gr 2)***
NERVOUS SYSTEM DISORDERS			
	Dizziness		
Dysgeusia			***Dysgeusia (Gr 2)***
	Headache		***Headache (Gr 3)***
		Leukoencephalopathy	
		Nervous system disorders - Other (cerebral infarction)	
	Paresthesia		
		Reversible posterior leukoencephalopathy syndrome	
		Transient ischemic attacks	
PSYCHIATRIC DISORDERS			
	Depression		
	Insomnia		***Insomnia (Gr 2)***
RENAL AND URINARY DISORDERS			
		Acute kidney injury	
		Proteinuria	
		Renal and urinary disorders - Other (nephrotic syndrome)	
RESPIRATORY, THORACIC AND MEDIASTINAL DISORDERS			
	Cough		***Cough (Gr 2)***

Adverse Events with Possible Relationship to Sunitinib malate (SU011248 L-malate) (CTCAE 4.0 Term) [n= 7115]			**Specific Protocol Exceptions to Expedited Reporting (SPEER)**
Likely (>20%)	**Less Likely (<=20%)**	**Rare but Serious (<3%)**	
	Dyspnea		***Dyspnea (Gr 3)***
	Epistaxis		***Epistaxis Gr 2)***
Laryngeal mucositis			***Laryngeal mucositis (Gr 2)***
Pharyngeal mucositis			***Pharyngeal mucositis (Gr 2)***
Tracheal mucositis			***Tracheal mucositis (Gr 2)***
SKIN AND SUBCUTANEOUS TISSUE DISORDERS			
	Alopecia		***Alopecia (Gr 2)***
	Dry skin		***Dry skin (Gr 2)***
		Erythema multiforme	
Palmar-plantar erythrodysesthesia syndrome			***Palmar-plantar erythrodysesthesia syndrome (Gr 3)***
	Pruritus		
	Rash maculo-papular		***Rash maculo-papular (Gr 3)***
	Skin and subcutaneous tissue disorders - Other (hair color change)		***Skin and subcutaneous tissue disorders - Other (hair color change) (Gr 2)***
		Skin and subcutaneous tissue disorders - Other (pyoderma gangrenosum)	
	Skin hypopigmentation		***Skin hypopigmentation (Gr 2)***
		Stevens-Johnson syndrome	
		Toxic epidermal necrolysis	
VASCULAR DISORDERS			
	Hypertension		***Hypertension (Gr 3)***
	Vascular disorders - Other (hemorrhage)(111)		

[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 perforation includes Colonic perforation, Duodenal perforation, Esophageal perforation, Gastric perforation, Ileal perforation, Jejunal perforation, Rectal perforation, and Small intestinal perforation under the GASTROINTESTINAL DISORDERS SOC.

[3]Allergic reactions observed include anaphylaxis and angioedema.

[4]The majority of hemorrhage events were mild. Major events, defined as symptomatic bleeding in a critical area or organ (e.g., eye, GI tract, GU system, respiratory tract, nervous system [including fatal intracranial hemorrhage, and cerebrovascular accident], and tumor site) have been reported.

Adverse events reported on Sunitinib malate (SU011248 L-malate) trials, but for which there is insufficient evidence to suggest that there was a reasonable possibility that Sunitinib malate (SU011248 L-malate) caused the adverse event:

BLOOD AND LYMPHATIC SYSTEM DISORDERS - Febrile neutropenia

CARDIAC DISORDERS - Atrial fibrillation; Cardiac arrest; Pericardial effusion

GASTROINTESTINAL DISORDERS - Ascites; Dysphagia; Gastrointestinal disorders - Other (enteritis); Hemorrhoids; Ileus; Small intestinal obstruction

GENERAL DISORDERS AND ADMINISTRATION SITE CONDITIONS - Pain

INVESTIGATIONS - GGT increased; INR increased

METABOLISM AND NUTRITION DISORDERS - Hypercalcemia; Hyperglycemia; Hyperkalemia; Hypocalcemia; Hypokalemia; Hyponatremia

MUSCULOSKELETAL AND CONNECTIVE TISSUE DISORDERS - Bone pain

NERVOUS SYSTEM DISORDERS - Cognitive disturbance; Nervous system disorders - Other (spinal cord compression); Peripheral sensory neuropathy; Seizure; Syncope

PSYCHIATRIC DISORDERS - Anxiety; Confusion

Renal and urinary disorders - Hematuria; Urinary retention

REPRODUCTIVE SYSTEMS AND BREAST DISORDERS - Hematosalpinx

RESPIRATORY, THORACIC AND MEDIASTINAL DISORDERS - Pharyngolaryngeal pain; Pleural effusion; Pneumothorax

VASCULAR DISORDERS - Flushing; Hypotension; Thromboembolic event

Note: Sunitinib malate (SU011248 L-malate) 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

7.2.1 CTCAE term (Adverse event 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. All appropriate treatment areas should have access to a copy of the CTCAE version 4.0. A copy of the CTCAE version 4.0 can be downloaded from the CTEP web site (http://ctep.cancer.gov).
"Expectedness":

Adverse events can be "Expected" (see Section 7.1) or unexpected. Bold and italicized terms in Section 7.1 identify expected events. See Section 7.5 for guidelines for reporting both types of events.

7.2.2 Attribution of the adverse event:

Definite – The adverse event is clearly related to the study treatment.

Probable – The adverse event is likely related to the study treatment.

Possible – The adverse event may be related to the study treatment.

Unlikely – The adverse event is doubtfully related to the study treatment.

Unrelated – The adverse event is clearly NOT related to the study treatment.

7.3 Expedited Adverse Event Reporting

Expedited adverse event reporting for this study is via CTEP-AERS (CTEP Adverse Event Reporting System), accessed via the secure CTEP web site https://eapps-ctep.nci.nih.gov/ctepaers/pages/task?rand=1397144505895). The reporting procedures to be followed are presented in the "NCI Guidelines: Expedited Adverse Event Reporting Requirements for NCI Investigational Agents" which can be downloaded from the CTEP web site (http://ctep.cancer.gov/protocolDevelopment/electronic_applications/docs/aeguidelines.pdf).

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, only when Internet connectivity is disrupted. Once Internet connectivity is restored, a 24-hour notification phoned in must be entered electronically into CTEP-AERS by the original submitter at the site.

CTEP-AERS is programmed for automatic electronic distribution of reports to the following individuals: Study Coordinator of the Lead Organization, Principal Investigator, and the local treating physician. CTEP-AERS provides a copy feature for other e-mail recipients.

7.3.1 Phase 1 and Early Phase 2 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

Note: All deaths on study require both routine and expedited reporting regardless of causality. Attribution to treatment or other cause must be provided.

- Expedited AE reporting timelines defined:
  - "24 hours; 5 calendar days" – The investigator must initially report the AE via CTEP-AERS within 24 hours of learning of the event followed by a complete CTEP-AERS report within 5 calendar days of the initial 24-hour report.
  - "10 calendar days" - A complete CTEP-AERS report on the AE must be submitted within 10 calendar days of the investigator learning of the event.

- Any medical event equivalent to CTCAE grade 3, 4, or 5 that precipitates hospitalization (or prolongation of existing hospitalization) must be reported regardless of attribution and designation as expected or unexpected with the exception of any events identified as protocol-specific expedited adverse event reporting exclusions.

- Any event that results in persistent or significant disabilities/incapacities, congenital anomalies, or birth defects must be reported via CTEP-AERS if the event occurs following treatment with an agent under a CTEP IND.

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

7.3.2 Expedited AE reporting timelines defined:

- "24 hours; 5 calendar days" – The investigator must initially report the AE via CTEP-AERS within 24 hours of learning of the event followed by a complete CTEP-AERS report within 5 calendar days of the initial 24-hour report.
- "10 calendar days" - A complete CTEP-AERS report on the AE must be submitted within 10 calendar days of the investigator learning of the event.

The Worldwide Product Safety (WPS) department at Merck will receive Adverse Experience reports, which are entered into the CTEP-AERS database, from the CTEP Contractor CTIS at the same time that they are forwarded to the NCI/CTEP.

Any medical event equivalent to CTCAE grade 3, 4, or 5 that precipitates hospitalization (or prolongation of existing hospitalization) must be reported regardless of attribution and designation as expected or unexpected with the exception of any events identified as protocol-specific expedited adverse event reporting exclusions.

Any event that results in persistent or significant disabilities/incapacities, congenital anomalies, or birth defects must be reported via CTEP-AERS if the event occurs following treatment with an agent under a CTEP IND.

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

7.3.3 Protocol-Specific Expedited Adverse Event Reporting Exclusions

For this protocol only, certain adverse events/grades are exceptions to the Expedited Reporting guidelines and do not require expedited reporting. The following adverse events should be reported through the routine reporting mechanism (Section 7.4):

CTCAE Category	Adverse Event	Grade	Hospitalization/ Prolongation of Hospitalization	Comments
GI disorders	Nausea	2 to 3		
	Diarrhea	2 to 3		
	Constipation	2 to 3		
	Vomiting	2 to 3		
Metabolism and nutrition disorders	Electrolyte abnormalities	2 to 4		
Nervous system disorders	Disguesia	2		
Dermatology/Skin	Rash Acne/Acneiform	2 to 3		

Events that are clearly consequences of the "main" event (e.g., hypokalemia associated with diarrhea or the arrhythmias, hypotension, hypoxia, etc. that are known to occur concurrently with

sepsis) may be noted in the Description of Event in the CTEP-AERS report and do not require separate CTEP-AERS reports.

The possibility of the contribution of comorbid conditions to the event should be considered when reporting adverse events. Examples include hyperglycemia in patients with diabetes or headaches and seizures in patients with brain tumors.

7.4 Routine Adverse Event Reporting

Those adverse events that do not require expedited reporting must be reported in routine (CTMS or CDUS; see Section 14.1) study data submissions. Adverse events reported through CTEP-AERS must also be reported in routine study data submissions.

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

8 NIH REPORTING REQUIREMENTS / DATA AND SAFETY MONITORING PLAN

8.1 Definitions

Please refer to definitions provided in Policy 801: Reporting Research Events found here.

8.2 OHSRP Office of Compliance and Training / IRB Reporting

8.2.1 Expedited Reporting

Please refer to the reporting requirements in Policy 801: Reporting Research Events and Policy 802 Non-Compliance Human Subjects Research found here.

8.2.2 IRB Requirements for PI Reporting at Continuing Review

Please refer to the reporting requirements in Policy 801: Reporting Research Events found here.

8.3 NCI Clinical Director Reporting

Problems expeditiously reported to the OHSRP/IRB in iRIS will also be reported to the NCI Clinical Director. A separate submission is not necessary as reports in iRIS will be available to the Clinical Director.

In addition to those reports, all deaths that occur within 30 days after receiving a research intervention should be reported via email to the Clinical Director unless they are due to progressive disease.

To report these deaths, please send an email describing the circumstances of the death to Dr. Dahut at NCICCRQA@mail.nih.gov within one business day of learning of the death.NIH Required Data and Safety Monitoring Plan

8.3.1 Principal Investigator/Research Team

The clinical research team will meet on a weekly basis when patients are being actively treated on the trial to discuss each patient. Decisions about dose level enrollment and dose escalation if applicable will be made based on the toxicity data from prior patients.

All data will be collected in a timely manner and reviewed by the principal investigator or a lead associate investigator. Events meeting requirements for expedited reporting as described in section **8.2.1** will be submitted within the appropriate timelines.

The principal investigator will review adverse event and response data on each patient to ensure safety and data accuracy. The principal investigator will personally conduct or supervise the investigation and provide appropriate delegation of responsibilities to other members of the research staff.

9 PHARMACEUTICAL INFORMATION

9.1 CTEP-Supplied Investigational Agent(s)

9.1.1 Erlotinib

OSI-774 (NSC 718781)

9.1.1.1 Chemical Name:

N-(3-ethynylphenyl)-6,7-bis(2-methoxyethoxy)-4-quinazolinamine, monohydrochloride

9.1.1.2 Other Names:

Erlotinib hydrochloride, Tarceva™

9.1.1.3 Classification:

Tyrosine kinase Inhibitor (EGFR)

9.1.1.4 Molecular Formula:

$C_{22}H_{23}N_3O_4 \cdot HCl$ M.W.: 393.4 (free base)

429.9 (hydrochloride salt)

9.1.1.5 Mode of Action:

Direct inhibition of EGFR tyrosine kinase

9.1.1.6 How Supplied:

Erlotinib tablets are provided by Astellas Pharma and distributed by the Pharmaceutical Management Branch, DCTD/NCI as 25 mg, 100 mg, and 150 mg white film-coated immediate-release tablets packaged in high-density polyethylene (HDPE) bottle. Each bottle contains 30 tablets.

The tablets are round and convex without markings. The 25 mg tablets are 1/4 inches (6 mm); the 100 mg tablets are 11/32 inches (9 mm); and the 150 mg tablets are 13/32 inches (10 mm). Erlotinib excipients include lactose monohydrate, hypromellose, hydroxypropyl cellulose, magnesium stearate, microcrystalline cellulose, sodium starch glycolate, sodium lauryl sulfate, and titanium dioxide. The tablets also contain trace amounts of color additives, including FD&C Yellow No. 6 (25 mg only) for product identification.

9.1.1.7 Storage:

Store at 25°C (77°F) in the original container; excursions permitted to 15° - 30°C (59° - 86°F).

9.1.1.8 Stability:

Current data indicates erlotinib is stable for at least 3 years at room temperature.

9.1.1.9 Route of Administration:

Oral.

9.1.1.10 Method of Administration:

- Tablets should be taken once daily preferably in the morning with up to 200 mL of water one hour before or two hours after food.
- Administration through G-tube: Dissolve the dose in 100 mL of sterile water, and shake it vigorously to form a uniform suspension. Draw suspension into a syringe and administer through the G-tube port. Repeat the transfer until the entire volume has been administered. Add small volume (40 mL) of sterile water to the container used to dissolve the tablets. Shake the residual suspension, aspirate it into a syringe, and administer. Repeat this last step to ensure the entire dose is administered. The total volume of delivery/rinse is ~180 mL.

9.1.1.11 Potential Drug Interaction:

Erlotinib is highly protein bound (92% to 95% in humans) and metabolizes primarily via CYP3A4 enzymes. Dose erlotinib cautiously with agents that are highly protein bound or potent CYP3A4 inhibitors/inducers enzymes.

CYP Iso-Enzymes Inhibitors/Inducers:

- Potent CYP3A4 Inhibitors: Use alternative drug. Alternatively, reduce erlotinib dose in the event of drug interaction (if permitted by the protocol).
- Potent CYP3A4 inducers: Use alternative drug. If an alternative treatment is contraindicated, consider increasing the erlotinib dose (if permitted by the protocol).
- Food-drug interaction: Avoid grapefruit /grapefruit juice (potent CYP3A4) while taking erlotinib.
- Smoking: Advise smokers to stop smoking while taking erlotinib. Smoking induces CYP1A2 enzymes and alters Erlotinib exposure by 64%.

Anticoagulant: Concomitant NSAIDs, warfarin or warfarin-derivatives may increase bleeding and prothrombin time (PT) / international normalized ratio (INR). Dose adjustment may be needed.

Patients receiving warfarin: Severe and fatal hemorrhage associated with elevated INRs can occur when OSI-774 and warfarin are administered concurrently. Patients taking warfarin or other coumarin-derivative anticoagulants should have more frequent INR/PT determinations (e.g., weekly for the first month and weekly for a minimum of 2 weeks following discontinuation of Erlotinib).

Statin: The combination of Erlotinib and a statin may increase the potential for statin-induced myopathy, including rhabdomyolysis, which was observed rarely.

Proton Pump Inhibitor: Erlotinib's solubility decreases as the pH increases. Co-administration of omeprazole with Erlotinib will increase the AUC and C_{max} by 46% and 61%, respectively.

H_2-antagonist: Avoid concomitant use of Erlotinib with gastric acid reducing agents if possible. When ranitidine 300 mg is given with Erlotinib, Erlotinib AUC and C_{max} decrease by 33% and 54%, respectively. Increasing the dose of Erlotinib will not compensate the loss of exposure. However, if an H_2-antagonist receptor is needed, **take Erlotinib at least 2 hours before or 10 hours following the H_2-antagonist administration**. Dosing such, Erlotinib loss of exposure is minimized to AUC of 15% and C_{max} of 17%.

Gastrointestinal perforation: Concomitant use of anti-angiogenic agents, corticosteroids, NSAIDs, and/or taxane based chemotherapy, or patients with prior medical history with peptic ulcers or diverticular disease are at high risk of GI perforation while on Erlotinib treatment. Discontinue Erlotinib if GI perforation manifests.

Patient Care Implications: If patient vomits after taking the tablets, readminister the dose only if the tablets can actually be seen and counted.

Recommend patients use sunscreen protection, and wear hat and long sleeve shirts as sunlight can exacerbate skin reactions.

9.1.1.12 Availability

Erlotinib is an investigational agent supplied to investigators by the Division of Cancer Treatment and Diagnosis (DCTD), National Cancer Institute (NCI).

Erlotinib is provided to the NCI under a Clinical Trials Agreement (CTA) between Astellas Pharma and the DCTD, NCI.

9.1.2 AZD6244

AZD6244 hydrogen sulfate (NSC 748727)

9.1.2.1 Chemical Name:

6-(4-Bromo-2-chloro-phenylamino)-7-fluoro-3-methyl-3H-benzoimidazole-5-carboxylic acid (2-hydroxy-ethoxy)-amide hydrogen sulfate

Other Names: ARRY-142886; AR00142866; AR-142886-01, AZD6244, AZD6244 Hyd-Sulfate,

9.1.2.2 Classification:

Mitogen-activated protein kinase (MEK) inhibitor

CAS Registry Number: 943332-08-9

9.1.2.3 Molecular Formula:

C17H15BrClFN4O3 · H2SO4 M.W.: 555.7

9.1.2.4 Mode of Action:

The RAS/RAF/MEK/ERK pathway is an important mediator of many cellular processes including proliferation, survival, differentiation, apoptosis, motility, and metabolism. This pathway is often aberrantly activated in human tumors due to the overexpression of mutant KRAS, mutant BRAF, or other growth factor receptors. AZD6244 is a selective mitogen-activated protein kinase (MEK) inhibitor. By inhibiting MEK, AZD6244 inhibits ERK

phosphorylation. Thus, AZD6244 may inhibit oncogenic growth signaling in tumor cells by targeting the RAS/RAF/MEK/ERK pathway.

9.1.2.5 How Supplied:

AZD6244 hydrogen sulfate is supplied as a 25 mg, size 4, plain, white, hydroxypropyl-methylcellulose (HPMC) capsule in white high density polyethylene (HDPE) containers with foil-lined, induction-sealed, child-resistant closures. Each bottle contains 60 capsules.

Each capsule contains a dispersion of AZD6244 hydrogen sulfate in d-α-tocopheryl polyethylene glycol 1000 succinate (TPGS; a water soluble form of vitamin E).

9.1.2.6 Storage:

Store the AZD6244 hydrogen sulfate capsules at room temperature (20°C-25°C). Brief excursions are permitted between 15°C and 30°C.

9.1.2.7 Stability:

Stability studies are ongoing.

9.1.2.8 Route of Administration:

Oral. Take AZD6244 on an empty stomach (either 1 hour before or 2 hours after meals). AZD6244 capsules should be taken with water only.

9.1.2.9 Potential Drug Interactions:

AZD6244 is primarily metabolized by CYP1A2 to N-desmethyl AZD6244 which is 3-5 fold more pharmacologically active than AZD6244. *In vitro*, AZD6244 is metabolized to a lesser extent by CYP2C19 and CYP3A4.

In vitro, AZD6244 is a weak inhibitor of CYP2C9 and CYP1A2.

High vitamin E doses may potentiate warfarin's anticoagulant activity. Monitor PT/INR more frequently in patients receiving both warfarin and AZD6244 hydrogen sulfate capsules.

Avoid concomitant intake of Vitamin E in excess of 100% of the recommended daily dose.

9.1.2.10 Availability:

AZD6244 is an investigational agent supplied to investigators by the Division of Cancer Treatment and Diagnosis (DCTD), NCI. AZD6244 is provided to the NCI under a Collaborative Agreement between AstraZeneca and the DCTD, NCI (see Section 14.3).

9.1.3 MK-2206

MK-2206 (NSC 749607)

9.1.3.1 Chemical Name or Amino Acid Sequence:

8-[4-(1-aminocyclobutyl)phenyl]-9-phenyl-1,2,4-triazolo[3,4-*f*]-1,6-naphthyridin-3(2H)-one mono-hydrochloride salt

9.1.3.2 Classification: Akt inhibitor

9.1.3.3 Molecular Formula:

C25H22N5OCl M.W.: 443.93

Approximate Solubility: Soluble in water (7.54 mg/mL; pH = 6.13) but less soluble in acetonitrile (1.4 mg/mL) and ethanol (2 mg/mL). Its mono-hydrochloride salt is slightly hygroscopic (absorbs 1.9 wt% water up to 95% relative humidity).

9.1.3.4 Mode of Action:

The PI3K-Akt pathway is activated downstream of EGFR, HER2, IGF1R, and cMet, and is a suspected driver of tumor progression in most cancers. Overexpression or activating mutations in receptor tyrosine kinases, PI3K and Ras, inactivation of the tumor suppressor PTEN, or amplification or mutation of Akt can activate Akt protein kinase in most carcinomas. It is believed that Akt inhibitors that target the pathway downstream of the most common mutations have broader utility and provide less resistance in the clinic.

9.1.3.5 Description:

A highly selective non-ATP competitive allosteric Akt inhibitor.

9.1.3.6 How Supplied:

MK-2206 tablets are supplied by Merck and distributed by the DCTD, NCI. The 5 mg, 25 mg, and 200 mg tablets are film coated, packaged in HDPE bottles. The 5 mg and 25 mg bottles contain 10 and 20 tablets, respectively. When inventory allows, the 5 mg and 25 mg bottles also contain 30 tablets each. The 200 mg bottles contain 5 tablets each. The pharmaceutical collaborator does not have stability data to support repackaging MK-2206 tablets in any container other than what is provided.

The white film (Opadry® 20A18273) coating consists of hydroxypropyl cellulose, hydroxypropyl methylcellulose and titanium dioxide.

Inactive ingredients consist of microcrystalline cellulose (Avicel PH102®), calcium phosphate dibasic anhydrous (ATAB®), croscarmellose sodium, and magnesium stearate.

9.1.3.7 Storage:

Store intact bottles at room temperature, not to exceed 30°C.

9.1.3.8 Stability:

Shelf life studies of MK-2206 are on going.

9.1.3.9 Route(s) of Administration: Oral

9.1.3.10 Method of Administration:

Take tablets 2 hours before or after food.

9.1.3.11 Potential Drug Interactions:

No clinical drug interaction studies have been performed with MK-2206. MK-2206 is not an inducer or inhibitor of major human P450 enzymes (CYP3A4, 2C9 and 2D6). In human hepatocytes, its metabolism involves both oxidation mainly by CYP3A4 and direct glucuronidation. MK-2206 is a substrate for P-glycoprotein (P-gp) mediated transport.

Patient Care Implications: In acute overdose, use activated charcoal to reduce the absorption of MK-2206. If additional measures are needed, consider emptying the stomach. Administer specific medical therapy as clinically appropriate.

9.1.4 Lapatinib

Lapatinib (NSC 727989)

9.1.4.1 Chemical Name:

N-{3-Chloro-4-[(3-fluorobenzyl)oxy]phenyl}-6-[5-({[2-(methylsulfonyl)ethyl]amino}methyl)-2 furyl]-4-quinazolinamine

Other Names: GW572016, Tykerb®

9.1.4.2 Molecular Formula:

C29H26ClFN4O4S(C7H8O3S)2H2O

Molecular Weight: 943.48

Approximate solubility: 0.007 mg/mL in water and 0.001 mg/mL in 0.1 N HCl at 25°C.

9.1.4.3 Mode of Action:

Dual inhibitor of epidermal growth factor receptor (EGFR or ErbB1) and ErbB2 tyrosine kinases.

9.1.4.4 How Supplied:

Lapatinib is supplied by the NCI/DCTD as 250 mg oval, biconvex, orange film-coated tablets with one side plain and the opposite side debossed with either FG HLS or GS XJG. The tablets contain 405 mg of lapatinib ditosylate monohydrate, equivalent to 250 mg lapatinib free base per tablet. The tablets are packaged into HDPE bottles with child-resistant closures containing 90 tablets per container.

Excipients present in the tablet include: Microcrystalline cellulose, povidone, sodium starch glycolate, and magnesium stearate.

The film-coat contains: Hydroxypropyl methylcellulose, titanium dioxide, macrogel/PEG 400, Polysorbate 80, FD&C Yellow No. 6, and FCF aluminum lake.

9.1.4.5 Storage:

Store intact bottles at controlled room temperature (15°C-30°C). Protect from light.

9.1.4.6 Stability:

Shelf life surveillance studies of the intact bottle are on-going. Current data indicates lapatinib is stable for at least 36 months at controlled room temperature (15°C - 30°C).

9.1.4.7 Route of Administration:

Oral on an empty stomach (either 1 hour before or 1 hour after meals).

9.1.4.8 Method of Administration:

Administer whole tablets. **Tablet crushing is not recommended.**

9.1.4.9 Potential Drug Interactions:

In vitro studies with human liver microsomes indicate that lapatinib is metabolized by CYP3A4 and CYP3A5, and to a lesser extent CYP2C19 and CYP2C8. Co-administration of lapatinib with potent or moderate CYP3A4 inhibitors (including grapefruit juice) and all CYP3A4

inducers is prohibited. Assess risk/benefit before co-administering lapatinib with weak CYP3A4 inhibitors. CYP3A4 inhibitors may decrease lapatinib metabolism (increasing levels); while CYP3A4 inducers may increase lapatinib metabolism (decreasing levels).

In human subjects, lapatinib inhibited CYP3A4 and CYP2C8 at clinically relevant concentrations. Avoid co-administration of lapatinib with drugs that are substrates of CYP3A4 or CYP2C8 and have narrow therapeutic windows.

Lapatinib potentially interacts with warfarin and quinazoline derivatives to increase INR and bleeding. Collect INR/PT determinations more frequently (e.g. weekly for the first month and weekly for a minimum of 2 weeks following lapatinib discontinuation).

9.2 Other Agent(s)

9.2.1 Sunitinib

Sunitinib malate (NSC 736511)

9.2.1.1 Chemical Name:

5-(5-fluoro-2-oxo-1,2-dihydro-indol-(3Z)-ylidenemethyl)-2,4-dimethyl-1H-pyrrole-3-carboxylic acid(2-diethylamino-ethyl)-amide; compound with (S)-2-hydroxy-succinic acid.

Other names: SU011248 L-Malate salt; SU010398; PHA-290940AD; Sutent; SU011248

9.2.1.2 Classification:

Receptor tyrosine kinase inhibitor (RTK)

9.2.1.3 Molecular formula:

C22H27FN4O2.C4H6O5 M.W.: 532.56 Daltons

9.2.1.4 Physical description:

Yellow to orange powder

Cas registry number: 341031-54-7

Aqueous solubility:

Solvent	Solubility (mg/mL)
0.1 M HCl	59.1
pH 4.5 buffer	25.4
pH 6.8 buffer	37.8
pH 7.5 buffer	0.05
in water	1.6

Solubility in various solvents:

Solvent	Solubility (mg/mL)
Acetonitrile	0.1
Dimethyl sulfoxide	92.9
Tetrahydrofuran	0.2
Methanol	1.5
Ethanol	0.3
1-Butanol	0.1

1-Butano:Water (80/20 v/v)	6.2
N,N-Dimethylacetamide	37
N,N-Dimethylformamide	18.4

9.2.1.5 Mode of Action:

Sunitinib malate is a receptor tyrosine kinase inhibitor involved in tumor proliferation and angiogenesis, specifically inhibiting platelet derived growth factor receptor, vascular endothelial growth factor receptor, stem cell factor receptor, Fms-like tyrosine kinase-3 receptor, and ret proto-oncogene.

9.2.1.6 How Supplied:

Sunitinib malate capsule is commercially available through Pfizer, Inc. and will be purchased by the participating institutions in the following strengths:

Capsules: 12.5 mg, 25 mg, and 50 mg capsules with mannitol, croscarmellose sodium, povidone, and magnesium stearate. Each opaque plastic bottle contains 30 capsules.

Capsule strength	Description
12.5 mg	Swedish Orange, Size 4 hard gelatin capsule
25 mg	Swedish Orange/Caramel, Size 3 hard gelatin capsule.
50 mg	Caramel, Size 2 hard gelatin capsule

9.2.1.7 Storage:

Store at controlled room temperature (15 to 30°C), and protect from light.

9.2.1.8 Stability:

The expiration date can be found on the commercially available supplies.

9.2.1.9 Route of Administration:

Oral. Sunitinib malate may be administered without regard to meals.

9.2.1.10 Potential Drug Interaction:

Sunitinib malate is metabolized primarily by liver enzymes, particularly CYP3A4. Dose reduction with the CYP3A4 inhibitors is recommended, based on clinical symptoms.

Concomitant treatment with dysrhythmic drugs, i.e., terfenadine, quinidine, procainamide, sotalol, probucol, bepridil, haloperidol, risperidone, and indapamide, is not recommended.

Patients Care Implications: A yellow discoloration of the skin area may result following direct contact with the capsules. Wash the exposed area with soap and water immediately.

9.3 Agent Ordering

NCI supplied agents may be requested by the Principal Investigator (or their authorized designee) at each participating institution. Pharmaceutical Management Branch (PMB) policy requires that agent be shipped directly to the institution where the patient is to be treated. PMB does not permit the transfer of agents between institutions (unless prior approval from PMB is obtained.) The CTEP assigned protocol number must be used for ordering all CTEP supplied investigational agents. The responsible investigator at each participating institution must be

registered with CTEP, DCTD through an annual submission of FDA Form 1572 (Statement of Investigator), Curriculum Vitae, Supplemental Investigator Data Form (IDF), 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 investigator at that institution.

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 < https://eapps-ctep.nci.nih.gov/OAOP/pages/login.jspx >. Access to OAOP requires the establishment of a CTEP Identity and Access Management (IAM) account < https://eapps-ctep.nci.nih.gov/iam/ > and the maintenance of an "active" account status and a "current" password. For questions about drug orders, transfers, returns, or accountability, call (240) 276-6575 Monday through Friday between 8:30 am and 4:30 pm (ET) or email PMBAfterHours@mail.nih.gov anytime.

9.4 Agent Accountability

Agent Inventory Records – The investigator, or a responsible party designated by the investigator, must maintain a careful record of the inventory and disposition of all agents received from DCTD using the NCI Drug Accountability Record Form (DARF). (See the CTEP home page at http://ctep.cancer.gov for the Procedures for Drug Accountability and Storage and to obtain a copy of the DARF and Clinical Drug Request form.)

10 CORRELATIVE / SPECIAL STUDIES

If enough material is available (after the molecular profiling analyses described in Section 5 have been performed), and the patient provides consent, the remainder paraffin embedded tissue and / or frozen samples or tumor DNA will be used for further molecular analysis.

10.1 Sample Storage, Tracking and Disposition

All research samples will be numerically coded prior to storage to maintain patient confidentiality. Subject identity is not provided to research laboratory personnel; i.e., samples are analyzed in a blinded fashion. Subjects will be given the option of consenting to future use of their research samples per the informed consent process with their option declared in the consent document. Biospecimens will be collected and processed using validated SOPs that will ensure both specimen quality and patient confidentiality pursuant to informed consent provisions.

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. Coded, linked labels 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 de-linked specimens will be shipped for analysis and/or storage. De-linked specimen labels will indicate: protocol number, order in which the patient was enrolled in the trial, type of sample, collection time, and total volume collected, as appropriate.

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 personnel only. The only patient information available in the inventory system will be the patient sex, diagnosis, and level of informed consent provided. SOPs ensure that any changes in the 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.

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). The PI will record any loss or unanticipated destruction of samples as a deviation. Reporting will be per the requirements of section **8.2**.

10.2 Large Scale Detection of Mutations in Candidate Genes

The remainder paraffin embedded tissue and / or frozen samples or tumor DNA, will be sent to one of the two following laboratories for large scale detection of mutations in candidate genes using next generation sequencing technologies:

Dr. Paul Meltzer
Attn. Dr. Keith Killian
Clinical Molecular Profiling Core lab
National Cancer Institute
Building 37, Room 6144
37 Convent Drive
Bethesda, MD 20892
Phone: 301-496-0999
E-Mail: killianj@mail.nih.gov

Dr. Robert Searless
Massively Parallel Sequencing Shared Resource
Oregon Health & Science University
Mailcode CH14G
3303 SW Bond Ave
Portland, OR 97239

10.2.1 Sample processing

Genomic DNA from histologically characterized tissue will be sequenced at loci encoding each candidate gene. DNA will be amplified using a PCR device and using exon specific primers. The PCR products will then be pooled and sequenced directly using a next generation sequencing device (i.e. Illumina Genome Analyzer). This will allow the parallel sequencing of the entire panel of candidate genes with rapid turnaround.

10.2.2 Gene sequencing

Somatic gene mutation analysis including but not limited to the following list of genes will be performed in the available tumor samples:

Approved Gene	Approved Gene Name	Symbol Location	Sequence Accession IDs	Previous Symbols	Aliases
AKT1	v-akt murine thymoma viral oncogene homolog 1	14q32.32-q32.33	M63167 NM_005163		RAC, PKB, PRKBA, AKT
AKT2	v-akt murine thymoma viral oncogene homolog 2	19q13.1-q13.2	NM_001626		
ALK	anaplastic lymphoma receptor tyrosine kinase	2q23	D45915 NM_004304		CD246
APC	adenomatous polyposis coli	5q21-q22	M74088 NM_000038		DP2, DP3, DP2.5
ART	anaplastic lymphoma receptor tyrosine kinase	2p23	D45915 NM_004304		CD246
ATM	ataxia telangiectasia mutated	11q22-q23	AB209133 NM_000051	ATA, ATDC, ATC, ATD	TEL1, TELO1
BCL2	B-cell CLL/lymphoma 2	18q21.3	M14745 NM_000633, NM_000657		Bcl-2
BMPR1A	bone morphogenetic protein receptor, type IA	10q22.3	BC028383 NM_004329	ACVRLK3	ALK3, CD292
BMPR1B	bone morphogenetic protein receptor, type IB	4q23-q24	D89675 NM_001203		ALK6, CDw293
BMPR2	bone morphogenetic protein receptor, type II (serine/threonine kinase)	2q33-q34	Z48923	PPH1	BRK-3, T-ALK, BMPR3, BMPR-II
BRAF	v-raf murine sarcoma viral oncogene homolog B1	7q34	M95712 NM_004333		BRAF1
CDKN2A	cyclin-dependent kinase inhibitor 2A (melanoma, p16, inhibits CDK4)	9p21	L27211 NM_000077	CDKN2, MLM	CDK4I, p16, INK4a, MTS1, CMM2, ARF, p19, p14, INK4, p16INK4a
CDKN2B	cyclin-dependent kinase inhibitor 2B (p15, inhibits CDK4)	9p21	AB060808 NM_004936		P15, MTS2, INK4B, TP15, CDK4I, p15INK4b
CDKN2C	cyclin-dependent kinase inhibitor 2C (p18, inhibits CDK4)	1p32.3	BC000598 NM_001262		INK4C, p18

CDH1	cadherin 1, type 1, E-cadherin (epithelial)	16q22.1	L08599 NM_004360	UVO	uvomorulin, CD324
CTNNB1	catenin (cadherin-associated protein), beta 1, 88kDa	3p21	X87838 NM_001098210	CTNNB	beta-catenin
EGFR	epidermal growth factor receptor (erythroblastic leukemia viral (v-erb-b) oncogene homolog, avian)	7p12	NM_005228	ERBB	ERBB1
EML4	echinoderm microtubule associated protein like 4	2p21	AF177377 NM_019063	C2orf2	ROPP120, ELP120
EPHA3	EPH receptor A3	3p11.2	M83941 NM_005233	ETK, ETK1, TYRO4	HEK, HEK4
EPHA5	EPH receptor A5	4q13.1	L36644 NM_004439		Hek7, TYRO4, CEK7, EHK1
EPHA7	EPH receptor A7	6q16.3	L36642		Hek11
ERBB2	v-erb-b2 erythroblastic leukemia viral oncogene homolog 2, neuro/glioblastoma derived oncogene homolog (avian)	17q11.2-q12	X03363	NGL	NEU, HER-2, CD340, HER2
ERBB3	v-erb-b2 erythroblastic leukemia viral oncogene homolog 3 (avian)	12q13	M34309	LCCS2	HER3
ERBB4	v-erb-a erythroblastic leukemia viral oncogene homolog 4 (avian)	2q33.3-q34	L07868 NM_001042599		
FAS	Fas (TNF receptor superfamily, member 6)	10q24.1	M67454	FAS1, APT1, TNFRSF6	CD95, APO-1
FGFR1	fibroblast growth factor receptor 1	8p12	M34185	FLT2, KAL2	H2, H3, H4, H5, CEK, FLG, BFGFR, N-SAM, CD331
FGFR2	fibroblast growth factor receptor 2	10q25.3-q26	AK026508 NM_022976, NM_000141	KGFR, BEK, CFD1, JWS	CEK3, TK14, TK25, ECT1, K-SAM, CD332
FGFR3	fibroblast growth factor receptor 3	4p16.3	M64347	ACH	CEK2, JTK4, CD333
FGFR4	fibroblast growth factor receptor 4	5q33-qter	AF202063		JTK2, CD334
FLT4	fms-related tyrosine kinase 4	5q34-q35	X68203		VEGFR3, PCL

FZD1	frizzled homolog 1 (Drosophila)	7q21	AB017363 NM_003505		DKFZp564G072
GNAS	GNAS complex locus	20q13.2-q13.3	M21142 NM_000516	GNAS1	NESP55, NESP, GNASXL, GPSA, SCG6
GSK3B	glycogen synthase kinase 3 beta	3q13.3	BC012760		
HIF1A	hypoxia inducible factor 1, alpha subunit (basic helix-loop-helix transcription factor)	14q23.2	U22431 NM_001530		MOP1, HIF-1alpha, PASD8, HIF1, bHLHe78
HRAS	v-Ha-ras Harvey rat sarcoma viral oncogene homolog	11p15.5	AJ437024 NM_176795	HRAS1	
IGF1R	insulin-like growth factor 1 receptor	15q26.3	M69229 NM_000875		JTK13, CD221, IGFIR, MGC18216, IGFR
INHBA	inhibin, beta A	7p15-p13			
KDR	kinase insert domain receptor (a type III receptor tyrosine kinase)	4q11-q12	AF035121		FLK1, VEGFR, VEGFR2, CD309
KIT	v-kit Hardy-Zuckerman 4 feline sarcoma viral oncogene homolog	4q11-q12	S67773	PBT	CD117, SCFR, C-Kit
KRAS	v-Ki-ras2 Kirsten rat sarcoma viral oncogene homolog	12p12.1	BC010502 NM_033360	KRAS2	KRAS1
LRP1B	low density lipoprotein-related protein 1B (deleted in tumors)	2q21.2	AF176832 NM_018557		LRP-DIT
LTK	leukocyte receptor tyrosine kinase	15q15.1-q21.1	D16105		TYK1
MAPK1	mitogen-activated protein kinase 1	22q11.2	M84489	PRKM2, PRKM1	ERK, ERK2, p41mapk, MAPK2
MAPK3	mitogen-activated protein kinase 3	16p11.2	M84490	PRKM3	ERK1, p44mapk, p44erk1
MAP2K1	mitogen-activated protein kinase kinase 1	15q22.1-q22.33	L11284	PRKMK1	MEK1, MAPKK1

MAP2K2	mitogen-activated protein kinase kinase 2	19p13.3	L11285	PRKMK2	MEK2
MET	met proto-oncogene (hepatocyte growth factor receptor)	7q31	M35073		HGFR, RCCP2
mTOR	mechanistic target of rapamycin (serine/threonine kinase)	1p36	L34075 NM_004958	FRAP, FRAP2, FRAP1	RAFT1, RAPT1, FLJ44809
NF1	neurofibromin 1	17q11.2	NM_000267		
NOTCH1	Notch homolog 1, translocation-associated (Drosophila)	9q34.3	AF308602 NM_017617	TAN1	
NRAS	neuroblastoma RAS viral (v-ras) oncogene homolog	1p13.2	BC005219 NM_002524		
NTRK1	neurotrophic tyrosine kinase, receptor, type 1	1q21-q22	Y09028 NM_002529		TRK, TRKA, MTC
NTRK2	neurotrophic tyrosine kinase, receptor, type 2	9q22.1	AF410902		TRKB
NTRK3	neurotrophic tyrosine kinase, receptor, type 3	15q24-q25	U05012		TRKC
PAK3	p21 protein (Cdc42/Rac)-activated kinase 3	Xq22.3	AF068864 NM_002578	MRX30, MRX47	hPAK3, bPAK
PDGFRA	platelet-derived growth factor receptor, alpha polypeptide	4q12	D50001 NM_006206		CD140a, PDGFR2
PIK3CA	phosphoinositide-3-kinase, catalytic, alpha polypeptide	3q26.3	NM_006218		p110-alpha, MGC142161, PI3K, MGC142163
PLK1	polo-like kinase 1 (Drosophila)	16p	NM_005030	PLK	
PRKDC	protein kinase, DNA-activated, catalytic polypeptide	8q11	NM_001081640	HYRC, HYRC1	DNPK1, p350, DNAPK, XRCC7, DNA-PKcs
PRKCG	protein kinase C, gamma	19q13.4	M13977 NM_002739	PKCG, SCA14	PKCC, MGC57564
PTCH1	patched homolog 1 (Drosophila)	9q22.1-q31	AI494442 NM_000264	NBCCS, PTCH	BCNS
PTEN	phosphatase and tensin homolog	10q23	U92436 NM_000314	BZS, MHAM	MMAC1, TEP1, PTEN1
PTPRD	protein tyrosine phosphatase, receptor type, D	9p24.1-p23	X54133		PTPD, HPTP

RB1	retinoblastoma 1	13q14.2	M15400	OSRC	RB
RHOA	ras homolog gene family, member A	3p21.3	BC001360 NM_001664	ARH12, ARHA	RhoA, Rho12, RHOH12
SMAD2	SMAD family member 2	18q21	U65019 NM_005901	MADH2	MADR2, JV18-1
SMAD4	SMAD family member 4	18q21.1	U44378 NM_005359	MADH4	DPC4
SLC38A3	solute carrier family 38, member 3	3p21.3	U49082 NM_006841		G17, SN1
SMG1	SMG1 homolog, phosphatidylinositol 3-kinase-related kinase (C. elegans)	16p12.3	AB061371 NM_015092		LIP, KIAA0421, ATX
SMO	smoothened homolog (Drosophila)	7q32.1	U84401 NM_005631	SMOH	
STK11	serine/threonine kinase 11	19p13.3	U63333 NM_000455		PJS, LKB1
STK33	serine/threonine kinase 33	11p15.3	AJ303380 NM_030906		
TGFB1	transforming growth factor, beta 1	19q13.1	X02812	TGFB, DPD1	CED, TGFbeta
TGFB2	transforming growth factor, beta 2	1q41	M19154 NM_003238		
TP53	tumor protein p53	17p13.1	AF307851 NM_000546		p53, LFS1
TSC1	tuberous sclerosis 1	9q34	AF013168	TSC	KIAA0243, LAM, hamartin
TSC2	tuberous sclerosis 2	16p13.3	AB014460 NM_000548	TSC4	tuberin, LAM
Wnt	wingless-type MMTV integration site family, member 1	12q13	X03072	INT1	
ZMYND10	zinc finger, MYND-type containing 10	3p21.3	U70824 NM_015896		BLU

10.3 Other molecular profiling analyses in tumor samples

If enough material is available, other molecular profiling analyses including but not limited to the following will be performed:

1) Comparative genomic hybridization (CGH)
2) FISH analysis of candidate genes
3) Methylation analysis
4) MicroRNAs
5) Gene expression arrays
6) Full tumor genome sequencing

10.4 Non-invasive biomarkers studies (Only for patients enrolled at the NCI intramural program)

Biomarkers, including prognostic and predictive factors, are widely studied in oncology and are important in anticancer drug development (112). As mentioned above, recurrent somatic gene mutations, such as EGFR, KRAS, p53, and LKB-1, have been detected in lung cancers (113). Some are important prognostic or predictive factors for treatment (18). With the emergence and progress of molecular targeted therapies, it is appealing to assess systematically the genetic background of lung cancer patients. Comprehensive evaluation of mutations, however, is often hampered by the small sample size of bioptic material or availability of cytologic material only. Also, repeat biopsy at relapse to study potential resistant mutations (e.g. EGFR T790M mutation) is technically challenging and potentially dangerous. Detecting mutant tumor genes from peripheral blood circulation represents a potential alternative (114). Besides the detection of circulating tumor cells (115), a technique that still needs optimization, tumor DNA in the circulation is potentially a simpler and more attractive source of information (116).

Techniques used nowadays for mutation detection, however, have several limitations. Traditional Sanger's method has relatively low sensitivity and specificity, especially when the tumor cells are contaminated with normal cells. Several real-time PCR based applications, such as scorpions-amplification refractory mutation system, are more sensitive (17, 18). The real-time PCR based applications, however, need designed primers or probes and can only detect pre-specified known mutations. On the other hand, none of these techniques can quantify the presence of mutations, which can be important to evaluate the dynamics of tumor burden after treatment or evaluate gene copy number changes (e.g. MET).

Invented initially for faster and more accurate genomic sequencing, next-generation sequencing (NGS) techniques, such as the Illumina genome analyzer, are applied to mutation discovery, DNA-protein interaction, mRNA expression, noncoding RNAs study, and metagenomics studies (113, 117). Through high-throughput sequence analysis and high coverage of depth, NGS may overcome the disadvantages mentioned above. NGS has been applied to sequence the cancer and leukemia genomes(118). NGS-based applications, RNA-seq and ChIP-seq, are superior to mRNA microarray and ChIP-on-chip technique, respectively, because they do not require prior knowledge of RNA and DNA sequences. It has been reported that NGS can detect EGFR mutations in malignant pleural effusion specimens with low tumor content and can measure the proportion of DNA molecules carrying the mutation as well (119). In summary, NGS could be an ideal platform to study genetic biomarkers of lung cancer in the circulation.

In addition, urine samples are also ideal sources of biomarker researching. Urine samples have been evaluated for diagnosis of bladder cancer (120, 121). Giving that nucleic acids fragments can pass glomerulus and enter urine(122), it has been reported to be feasible to detect mutant

genes from urine in patients with non-genitourinary origin solid tumors(123). Because microRNAs are potential important biomarkers of lung cancer(124, 125), are small in size (22-26bp) to pass glomerulus, and are reported to be detectable in urine(126), it is interesting and reasonable to assess the role of urinary microRNA expression in lung cancer patients.

10.4.1 Blood sample analyses.

If possible, blood will be collected in one EDTA tube (5ml purple top) and one Sodium Heparin Tube (10ml green top) upon enrollment in the trial. The samples will be stored at 4°C and sent to the following laboratory(s) for analyses:

Dr. Udayan Guha

Attn. Dr. Steven Lee
National Cancer Institute
Building 10, Room 8N258
10 Center Drive
Bethesda, MD 20892
Phone: 301.402.0082

The blood will be centrifuged at 820g for 10 minutes. The plasma will be collected and stored in -80°C for further analyses that will include but are not limited to the following:

Circulating DNA mutation detection. The buffy coat, in which white blood cells and circulating tumor cells are located, will be isolated for possible circulating tumor cells analysis. DNA in the buffy coat will be extracted within 24 hours of blood sampling if no more analysis of circulating cells will be conducted.

DNA will be extracted from 1 ml of plasma. A pilot study, which will enroll around 10 patients, will be conducted to evaluate the feasibility of detecting mutant genes in circulating tumor DNA. In the pilot study, mutant genes confirmed in tumor samples will be analyzed in the plasma by an Ilumina Genome Analyzer. If mutant genes are detected, the amount of mutant gene will be represented by the ratio of mutant gene over wild-type gene, adjusted by the copy number of a reference gene, such as LINE-1 or hTERT gene.

After the pilot study and adjustment of experiments' conditions, circulating mutant genes will be measured regularly and their levels will be correlated with clinical treatment outcome. Screening of new mutations will be conducted if disease progression develops and its result will be compared with the result of tumor re-biopsy, if any.

10.4.2 Urine sample analyses.

If possible, 5 ml of urine will be collected in a yellow top urine tube upon enrollment in the trial. The container will be stored at 4°C and sent to the following laboratory(s) for analyses:

Dr. Udayan Guha

Attn. Dr. Steven Lee
National Cancer Institute
Building 10, Room 8N258
10 Center Drive
Bethesda, MD 20892
Phone: 301.402.0082

The urine will be used for further analyses that will include but are not limited to the following:

Urine micro RNA detection. Urine will be stored at 4°C and processed within 4 hours of collection. Urine will be mixed 1:1 with lysis buffer and frozen at -80°C for later DNA and RNA extraction. Urine RNA will be extracted. A panel of microRNA will be analyzed by real-time PCR, including let7a, mir21, mir29b, mir34a, mir34b, mir34c, and mir155. U6 snRNA will be measured as internal control. The association of microRNA results with patients' clinical characteristics and treatment outcomes will be examined.

10.4.3 Genomic Analysis

Whole exome, transcriptome, and whole genome (only on a subset of patients) sequencing will be performed to identify somatic changes in tumor tissue if tissue is available after the other studies mentioned above are performed. Comparative genomic hybridization (CGH) analyses will be performed to interrogate the copy number gains and losses. DNA and RNA will be extracted from tumor samples and germ line DNA and RNA will be extracted from peripheral blood mononuclear cells from whole blood.

DNA isolated from blood will be analyzed to identify the somatic and germline variants. Transcriptome sequencing will reveal expression at the RNA-level. The genomic analyses will be performed in ATC genome core facility, and in the laboratories of Drs. Meltzer, Raffeld and Guha.

10.4.3.1.1 DNA Sequencing

Whole-exome multiregion spatial sequencing of DNA will be performed using a next-generation sequencing device and somatic mutations identified will be validated and verified using Sanger sequencing (NCI Frederick sequencing facility). Whole genome sequencing will be performed on a subset of samples based on the results of whole-exome and transcriptome sequencing.

10.4.3.1.2 RNA Sequencing

Transcriptome profiling will be performed using RNA-Seq approach to study transcriptome complexity, and for identification of genes, structure of transcripts, alternative splicing, non-coding RNAs, and new transcription units (NCI Frederick sequencing facility). RNA-seq will also reveal expression of individual genes at the transcript-level. This will be correlated with quantitative protein expression by mass spectrometry.

10.5 Samples for Genetic/Genomic Analysis

10.5.1 Description of how privacy and confidentiality of medical information/biological specimens will be maximized

Tissue and blood samples will be coded. DNA, RNA and protein isolated from these tissues and cell lines and xenografts generated will all be similarly coded. Only personnel involved in this study will have access to both the code and the name of the patient. To help protect privacy, a Certificate of Confidentiality has been obtained from the National Institutes of Health. To facilitate genetic research, and for the purpose of publication of research work, data from genomic and proteomic studies may be deposited in appropriate public databases. Coded data will be deposited in a manner that the patient's identity cannot be traced.

10.5.2 Management of Results

Participants will be given the choice of whether or not they would like the Durable Power of Attorney (DPA) to receive notification if a gene variant known to cause or contribute to disease is identified. In cases where in the opinion of the principal investigator there is a finding of urgent importance to the health of the family members of the participants, the investigator will consult with an interdisciplinary group of geneticists, clinicians, and the NIH ethics board to determine whether the result needs to be divulged to immediate family members and the DPA will be notified by telephone or by mail. Attempts will be made to verify the results in a CLIA certified lab prior to notification. Results may become available for a period of up to two years.

10.5.3 Genetic Counseling

Genetic counseling will be provided to the patient by genetic counseling services of the Clinical Center upon consent to the protocol and during one of the routine follow up visits to the clinical center. In the event a result of urgent importance to immediate family members has to be divulged, family members will be offered genetic counseling.

11 DATA COLLECTION AND EVALUATION

11.1 Data Collection

The PI will be responsible for overseeing entry of data into in-house password protected electronic systems, C3D and LabMatrix, and ensuring data accuracy, consistency and timeliness. The principal investigator, associate investigators/research nurses and/or a contracted data manager will assist with the data management efforts. All data obtained during the conduct of the protocol will be kept in secure network drives or in approved alternative sites that comply with NIH security standards. Primary and final analyzed data will have identifiers so that research data can be attributed to an individual human subject participant.

All adverse events, including clinically significant abnormal findings on laboratory evaluations, regardless of severity, will be followed until return to baseline or stabilization of event.

Document AEs from the first study intervention through 30 days after the last study intervention. Beyond 30 days after the last intervention, only adverse events which are serious and related to the study intervention need to be recorded.

End of study procedures: Data will be stored according to HHS, FDA regulations, and NIH Intramural Records Retention Schecule as applicable.

Loss or destruction of data: Should we become aware that a major breach in our plan to protect subject confidentiality and trial data has occurred, this will be reported expeditiously per requirements in section **8.2.1**.

11.2 Genomic Data Sharing Plan

Unlinked genomic data will be deposited in public genomic databases such as dbGaP in compliance with the NIH Genomic Data Sharing Policy.

12 STUDY CALENDAR

Baseline evaluations are to be conducted within 1 week prior to start of protocol therapy. Scans and x-rays must be done <4 weeks prior to the start of 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. The study calendar does not apply to individuals enrolled for *EGFR* germline mutation testing only; please refer to Section 5.6 for testing and follow-up of these individuals.

		Screening – Part 1 Molecular Profiling and Correlative Studies	Screening – Part 2 Treatment Arm Eligibility (see Section 3.3)	Treatment Cycles (see Section 5.3; clinic visits may be extended to every 6 weeks for the Erlotinib, AZD6244, and Lapatinib arms; every 8 weeks for the MK-2206 arm; and every 12 weeks for the Sunitinib arm)	Off treatment
Biopsy for molecular profiling[11]		X		Upon disease progression per section 5 (optional)	
Targeted therapies[9]				See section 5 for details regarding agent administration and scheduling	
Informed consent		X		N/A	
Demographics		X			
NOS arm data[10]			X		
Medical history[9]			X	Every clinic visit	X
Concurrent Meds[9]			X	Every clinic visit	X
NIH Advance Directives Form[14]		X			
Physical exam[9]			X	Every clinic visit	X
Vital signs[9]			X	Every clinic visit	X
Height[9]			X	N/A	
Weight[9]			X	Every clinic visit	X
Performance status[9]			X	Every clinic visit	X
CBC w/diff, plts[9]			X	Prior to each clinic visit	X
Serum chemistry[1, 9]			X	Prior to each clinic visit	X
PT, aPTT[9]			X	As clinically indicated	X
Urinalysis[9]			X		X
EKG[9]			X		X
BP[3, 9]				Weekly during cycle 1	
ECHO/MUGA[4, 5, 9]			X	Every other treatment cycle	
Adverse event evaluation[9]				X----------X	X
Tumor measurements[9]			X	Approximately every 6, 8, or 12 weeks, as defined for each treatment arm. See Section 5.8.2 for details. Documentation (radiologic) must be provided for patients removed from study arm for progressive disease.	X

		Screening – Part 1 Molecular Profiling and Correlative Studies	Screening – Part 2 Treatment Arm Eligibility (see Section 3.3)	Treatment Cycles (see Section 5.3; clinic visits may be extended to every 6 weeks for the Erlotinib, AZD6244, and Lapatinib arms; every 8 weeks for the MK-2206 arm; and every 12 weeks for the Sunitinib arm)	Off treatment
Radiologic evaluation[9]			X	Approximately every 6, 8, or 12 weeks, as defined for each treatment arm. See Section 5.8.2 for details.	X
B-HCG[2, 9]			X	As clinically indicated	
$HgbA1_c$ [6, 9]				X----------X	
Correlative studies[7, 8, 9, 12]		X		Upon disease progression if repeat biopsy is performed	
EGFR germline mutation testing[13]		X			

[1]:Sodium, potassium, chloride, CO2, BUN, creatinine, glucose, AST, ALT, alkaline phosphatase, bilirubin, albumin, total protein, LDH, calcium, phosphorous and magnesium.
[2]:Serum pregnancy test (women of childbearing potential).
[3]: Required only for patients enrolled on a Sunitinib treatment arm, per section 6.2.1.
[4]: Required only for patients enrolled on a Lapatinib treatment arm, per section 6.1.5.
[5]: Required for patients enrolled on a Sunitinib treatment arm who have known cardiac dysfunction at study entry and/or clinically observed adverse cardiac events.
[6]: For patients enrolled on the MK-2206 arm, who require treatment of hyperglycemia per section 6.1.2, $HgbA1_c$ monitoring should be performed with each cycle of MK-2206 therapy
[7]: 1 5ml EDTA tube (purple top) and 1 urine sample collected in a (yellow top) urine tube. Page Betsy Morrow 10063 for pick up.
[8]: 1 10ml Sodium Heparin Tube (green top). Call Zied Abdullaev for pick up at 301 451-2711.
[9]: N/A for patients assigned to the NOS arm.
[10]: Required only for the patients assigned to the NOS arm.
[11]: Including any other procedures necessary to do the biopsy, as clinically indicated.
[12]: Correlative Studies: See Section 9 for details.
[13]: Selected patients only (see Section 5.6). Patient must meet eligibility requirements for *EGFR* germline mutation testing and have signed a separate consent form.
[14]: As indicated in section **16.3**, all subjects ≥ age 18 will be offered the opportunity to complete an NIH advance directives form. This should be done preferably at baseline but can be done at any time during the study as long as the capacity to do so is retained. The completion of the form is strongly recommended, but is not required.

13 MEASUREMENT OF EFFECT

13.1 Antitumor Effect – Solid Tumors

For the purposes of this study, patients should be re-evaluated for response every 6 +/- 1 weeks. In addition to a baseline scan, confirmatory scans should also be obtained within 4 to 6 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.

13.1.1 Definitions

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

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.

13.1.2 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, as >10 mm with CT scan, or >10 mm with calipers by clinical exam. All tumor measurements must be recorded in millimeters (or decimal fractions of centimeters).

Note: Tumor lesions that are situated in a previously irradiated area will not be considered measurable.

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.

13.1.3 Methods 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).

Use of MRI remains a complex issue. MRI has excellent contrast, spatial, and temporal resolution; however, there are many image acquisition variables involved in MRI, which greatly impact image quality, lesion conspicuity, and measurement. Furthermore, the availability of MRI is variable globally. 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:

- Negative FDG-PET at baseline, with a positive FDG-PET at follow-up is a sign of PD based on a new lesion.
- 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.
- 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. However, it must be acknowledged that both approaches may lead to false positive CR due to limitations of FDG-PET and biopsy resolution/sensitivity. In addition, the FDG-PET may not be used to upgrade a response to a CR if the patient is actively being maintained on MK-2206, since the AKT inhibitor blocks intracellular glucose transport and may cause a negative FDG-PET result as an artifact, due to its pharmacodynamic target effect.

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.

13.1.4 Response Criteria

13.1.4.1 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

13.1.4.2 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).

13.1.4.3 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

13.1.5 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.

13.1.6 Progression-Free Survival

PFS is defined as the duration of time from start of treatment to time of progression or death, whichever occurs first.

14 DATA REPORTING / REGULATORY REQUIREMENTS

Adverse event lists, guidelines, and instructions for adverse event reporting can be found in Section **7.2** (Adverse Events: List and Reporting Requirements).

14.1 Data Reporting

14.1.1 Method

This study will be monitored by the Clinical Data Update System (CDUS) version 3.0. Cumulative CDUS data will be submitted quarterly to CTEP by electronic means. Reports are due January 31, April 30, July 31 and October 31. Instructions for submitting data using the CDUS can be found on the CTEP web site (http://ctep.cancer.gov/reporting/cdus.html).

14.1.2 Responsibility for Data Submission

Study participants are responsible for submitting CDUS data and/or data forms to the Coordinating Center quarterly at least 2 weeks before CTEP's due date to allow time for Coordinating Center compilation, Principal Investigator review, and timely submission to CTEP (see Section **14.1.1**). The Coordinating Center is responsible for compiling and submitting CDUS data to CTEP for all participants and for providing the data to the Principal Investigator for review.

14.2 CTEP Multicenter Guidelines

This protocol will adhere to the policies and requirements of the CTEP Multicenter Guidelines. The specific responsibilities of the Principal Investigator and the Coordinating Center (Study Coordinator) and the procedures for auditing are presented in Appendix I.

The Principal Investigator/Coordinating Center is responsible for distributing all IND Action Letters or Safety Reports received from CTEP to all participating institutions for submission to their individual IRBs for action as required.

Except in very unusual circumstances, each participating institution will order DCTD-supplied agents directly from CTEP. Agents may be ordered by a participating site only after the initial IRB approval for the site has been forwarded by the Coordinating Center to the CTEP PIO (PIO@ctep.nci.nih.gov) except for Group studies.

14.3 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, Agent-CRADA, CTA, CSA) between the Pharmaceutical Company(ies) (hereinafter referred to as a "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/default.htm) contained within the terms of award, apply to the use of the Agent(s) in this study:

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 investigational 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.

2. For a clinical protocol where there is an investigational Agent used in combination with (an)other investigational 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"):

a. NCI will provide all Collaborators with prior written notice regarding the existence and nature of any agreements governing their collaboration with NIH, 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 investigational 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 investigational Agent.

3. Clinical Trial Data and Results and Raw Data developed under a Collaborative Agreement will be made available exclusively to Collaborator(s), the NCI, and the FDA, as appropriate and unless additional disclosure is required by law or court order. 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.

4. 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.

5. 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.

6. 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:

Jan M. Casadei, Ph.D.,
Chief, Regulatory Affairs Branch
Cancer Therapy Evaluation Program
Division of Cancer Treatment and Diagnosis, NCI
9609 Medical Center Drive
Room 5-W532, MSC 9740
Bethesda, MD 20892-9740 [if US Postal Service]
Rockville, MD 20850 [if non-USPS/private carrier]
Tel. 240-276-6125
E-mail: casadeij@mail.nih.gov
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.

15 STATISTICAL CONSIDERATIONS

15.1 Study Design/Endpoints

The primary objectives of this study are to determine the feasibility and efficacy of the use of molecular profile directed therapies in the treatment of advanced thoracic malignancies. The feasibility rate for the trial will be evaluated by determining the percentage of enrolled patients with a successful molecular profile determined. Efficacy will be determined by assessing if patients who have treatment assigned on the basis of their molecular profiling results will exhibit reasonable response rates to the drug selected for their particular profile.

Patients with NSCLC, SCLC, and thymic malignancies will be eligible for enrollment and each will be considered a separate cohort. In addition, on the basis of the molecular profiling results, one of 5 specific treatments could be assigned to each patient. The particular treatment assignments will be made according to a cascading schema as shown. If no profile of one of the 5 types listed is identified as being appropriate for a given patient, patients will be followed and treated in a separate 'other' (NOS) category.

With 3 disease types and 5 possible specific treatments for each, there are 15 possible treatment arms which will be under active consideration. For each of these 15, the study will be conducted as an optimal two-stage phase II trial (Simon R, Controlled Clinical Trials 10:1-10, 1989).

In all but the arm for patients who have NSCLC and an EGFR mutation, the trial will be conducted in order to rule out an unacceptably low 10% clinical response rate (PR+CR; p0=0.10) in favor of a modestly high response rate of 40% (p1=0.40). For those 14 arms, with alpha=0.05 (probability of accepting a poor treatment=0.05) and beta = 0.10 (probability of rejecting a good treatment=0.10), the study will initially enroll 9 evaluable patients in each stratum and if 0 or 1 of the 9 have a clinical response, then no further patients will be accrued. If 2 or more of the first 9 have a response, then accrual would continue until a total of 20 patients have enrolled in that stratum. If there are 2-4 responses in 20 patients, this would be an uninterestingly low response rate in that stratum, while if there were 5 or more responses in 20 patients, then this would be sufficiently interesting to warrant further study in later trials. Under the null hypothesis (10% response rate), the probability of early termination is 77% for a given stratum.

In the arm for patients who have NSCLC and an EGFR mutation, the trial will be conducted in order to rule out an unacceptably low 30% clinical response rate (PR+CR; p0=0.30) in favor of a modestly high response rate of 60% (p1=0.60). The target is higher for these patients since a higher response rate is known to be possible for these patients. For that one arm, with alpha=0.05 (probability of accepting a poor treatment=0.05) and beta = 0.10 (probability of rejecting a good treatment=0.10), the study will initially enroll 10 evaluable patients in each stratum and if 0 to 3 of the 10 have a clinical response, then no further patients will be accrued. If 4 or more of the first 10 have a response, then accrual would continue until a total of 28 patients have enrolled in that stratum. Under the null hypothesis (30% response rate), the probability of early termination is 65% for patients in this stratum.

As feasibility is an important endpoint as well, the trial will include an early stopping rule for accrual to specific arms which are not enrolling adequate numbers of patients. After 18 months from the date the first patient enrolls on the trial in any arm, if there are arms with no more than a

single patient, then further accrual to that arm will end. This will prevent continued accrual to arms which are very unlikely to be able to treat sufficient patients for evaluating the first stage (e.g., 9 or 10 patients) within 5 years.

15.2 Sample Size/Accrual Rate

With a maximum of 20 patients per stratum, up to 300 evaluable patients may be treated in specific arms. All patients in the 'other' (NOS) stratum will be evaluated and have their data analyzed by disease and type of treatment to the extent possible. As they are considered entirely exploratory, no specific targets for accrual will be used for the patients in the 'other' (NOS) stratum. For the secondary objective of determining the frequency of *EGFR* germline mutations in families with high susceptibility to lung cancer, we plan to enroll 15 families over a period of a year; however, specific accrual targets are not provided due to the exploratory nature of this endpoint.

In order to enroll up to 300 patients in the specific arms on the basis of molecular profiling results, it is anticipated that based on the rates of mutations that occur in the genes used for treatment assignments, approximately 600 patients may need to be enrolled onto the trial. It is anticipated that up to 10 patients per month may be enrolled onto this trial from all participating sites and in all diseases combined, and thus approximately five years may be required to enroll up to 600 patients. Far fewer patients may be enrolled if the treatments fail to allow accrual to the second stage of the trial or if some arms close to accrual due to feasibility issues.

15.3 Stratification Factors

N/A

15.4 Analysis of Secondary Endpoints

In order to accomplish the secondary objective of identifying molecular profiles in patients with NSCLC, SCLC or thymic malignancies and characterize their natural histories, clinical course and response to treatment, we will follow all patients that undergo molecular profiling from the time that they are enrolled in the trial until the time of death. Demographic information such as age, gender and race will be tabulated. The frequencies of oncogenic mutations will be assessed. The exact binomial two-sided 95% confidence interval for each mutation rate will be reported.

Kaplan-Meier curves for progression free survival, duration of response, and overall survival will be calculated from the patients treated in the different treatment arms of this study (erlotinib, lapatinib, sunitinib, AZD6244, MK2206, or NOS). The results of this analysis may be compared in an informal manner to any similarly defined curves available from other published studies in comparable patients with the same disease. Expression levels of various proteins from pre- and post-treatment samples will be compared using an appropriate nonparametric test and pre-post differences between responders and non-responders using a Wilcoxon rank sum test. These latter evaluations will be considered exploratory and the resulting p-values will be presented as being exploratory and without adjustment for multiple comparisons.

For the secondary objective of determining the frequency of *EGFR* germline mutations in families with high susceptibility to lung cancer, the analyses will be performed in an exploratory manner. Hence, statistical power calculations have not been provided.

15.5 Reporting and Exclusions

15.5.1 Evaluation of toxicity

All patients will be evaluable for toxicity from the time of the biopsy for molecular profiling is performed. Patients who are subsequently enrolled in the NOS arm will not be evaluable for toxicity.

15.5.2 Evaluation of response

All patients included in one of the experimental arms of the study must be assessed for response to treatment, even if there are major protocol treatment deviations. 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 for a particular treatment arm (with the exception of those who received no study medication) should be included in the main analysis of the response rate. Patients in response categories 5-9 should 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 5-9 will be protocol specific.

All conclusions should be based on all eligible patients. Subanalyses 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 deviations, etc.). However, these subanalyses may not serve as the basis for drawing conclusions concerning treatment efficacy, and the reasons for excluding patients from the analysis should be clearly reported. The 95% confidence intervals should also be provided.

16 HUMAN SUBJECTS PROTECTIONS

16.1 Rationale for Subject Selection

This study will be open to all individuals with advanced thoracic malignancies (lung cancer and thymic malignancies) regardless of gender, ethnicity, or race provided that the aforementioned inclusion and exclusion criteria are met. For safety reasons, only pregnant women and children are excluded from this study. This study will be recruited through internal referral, our local physician referral base, and through various cancer information hotlines (i.e., Clinical Studies Support Center, 1-800-4Cancer). This is a trial designed to evaluate the efficacy of different targeted therapies in patients with advanced thoracic malignancies. Patients should realize that we are hopeful that they may gain benefit from this study, but there is no objective evidence to support our optimism at this time. Subjects from both genders and all racial /ethnic groups are eligible for this study if they meet the eligibility criteria outlined in Section 3. To date, there is no information that suggests that differences in drug metabolism or disease response would be expected in one ethnic group compared to another. Efforts will be made to extend accrual to each representative population, but in this preliminary study, a balance must be struck between

patient safety considerations and limitations on the number of individuals exposed to potentially toxic and/or 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, accrual may be expanded or a follow-up study may be written to investigate those differences more fully.

16.2 Justification for Exclusions

Due to lack of knowledge of the effects of most or all of the targeted agents included in this trial on the fetus or on 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 (ongoing or active infection, symptomatic congestive heart failure (AHA Class II or worse), unstable angina pectoris, cardiac arrhythmia, or psychiatric illness/social situations that would limit compliance with study requirements) are excluded due to the possibility that the agents used may worsen their condition and the likelihood that the underlying condition may obscure the attribution of adverse events with respect to protocol therapy.

16.2.1 Participation of Children

Patients under the age of 18 will be excluded from study due to the low occurrence of these oncologic histologies in the pediatric population. In addition, the risk of exposure to an investigational agent without proven benefit in the targeted histologies supports excluding children until additional safety and efficacy data is available. Patients under the age of 18 will be excluded from germline mutation testing because of the infrequency of lung cancer in this age group and because the results would not influence the medical management of individuals under the age of 18.

16.3 Participation of Subjects Unable to Give Consent

Adults unable to give consent are excluded from enrolling in the protocol. However, re-consent may be necessary and there is a possibility, though unlikely, that subjects could become decisionally impaired. For this reason and because there is a prospect of direct benefit from research participation (section **16.4**), all subjects ≥ age 18 will be offered the opportunity to fill in their wishes for research and care, and 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 in the event that they become incapacitated or cognitively impaired during the course of the study. Note: The PI or AI will contact the NIH Ability to Consent Assessment Team (ACAT) for evaluation as needed for the following: an independent assessment of whether an individual has the capacity to provide consent; assistance in identifying and assessing an appropriate surrogate when indicated; and/or an assessment of the capacity to appoint a surrogate. For those subjects that become incapacitated and do not have pre-determined substitute decision maker, the procedures described in NIH HRPP SOP 14E for appointing a surrogate decision maker for adult subjects who are (a) decisionally impaired, and (b) who do not have a legal guardian or durable power of attorney, will be followed.

16.4 Evaluation of Benefits and Risks/Discomforts

The potential benefit to a patient who enters study is a reduction in the bulk of his/her tumor, which may or may not have a favorable impact on symptoms and/or survival. Potential risks

include the possible occurrence of any of a range of side effects that are listed in the pharmaceutical section and the consent document. The procedure for protecting against or minimizing risks will be to medically evaluate patients on a regular basis as described earlier.

As of 3/22/2016, the treatment arms of this study have been closed. The study remains open under NIH Intramural IRB for the molecular profiling arm only. Participating in the molecular profiling arm is associated with risks that may accompany the biopsy required for genetic testing. These risks include bleeding or infection at the biopsy site as well as potential radiation exposure if the biopsy is done with CT guidance. Additionally, participation comes with the risk of discovering new genetic results that may have implications on the participant and/or family members. The benefits of participation include gaining additional genetic knowledge which may provide guidance in cancer treatment options provided outside of this study. Pre and post testing genetic counseling will be provided as these results are disclosed to provide assistance in the handling of these results.

16.5 Risk/Benefit Analysis

As of 3/22/2016, the treatment arms of this study have been closed, but study remains open under NIH Intramural IRB for molecular profiling arm only. For adults in the molecular profiling arm, including those who become unable to consent, there is more than minimial risk in participation associated with the biopsy with the possibility of direct benefit by way of return of genetic findings that may provide additional treatment options outside of this study.

16.6 Consent and Assent Process and Documentation

An associate or principal investigator on the trial will inform patients of the purpose, alternatives, treatment 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 at a subsequent visit. The original of the signed informed consent will be placed in the patient's medical record and a copy will be held 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. The proband as well as all family members undergoing germline genetic testing will sign a separate informed consent for germline mutation testing. For deceased family members of the proband with history of lung cancer, who have available tissue, consent will be obtained from the next of kin.

16.6.1 Telephone Consent (for amendments only):

The informed consent document will be sent to the subject. An explanation of the study will be provided over the telephone after the subject has had the opportunity to read the consent form. The subject will sign and date the informed consent.

The original informed consent document will be sent back to the consenting investigator who will sign and date the consent form with the date the consent was obtained via telephone.

A fully executed copy will be returned via mail for the subject's records.

The informed consent process will be documented in the medical record.

16.7 DATA AND SAFETY MONITORING PLAN

Any new significant finding that may affect the patient's willingness to continue in the study will be shared with patients. Data will be monitored regularly by the principal investigator in order to identify significant toxicity trends. Confidentiality will be maintained as much as possible, consistent with applicable regulations. Names of participants or identifying material will not be released without patient permission, except when such release is required by law. No patient's name or identifying information will be released in any publication or presentation. Records are maintained according to current legal requirements and are made available for review according to the requirements of the Food and Drug Administration (FDA) or other authorized user, only under guidelines established by the Federal Privacy Act.

This study will also be monitored by the National Cancer Institute Safety Monitoring Committee (SMC). The Coordinating Center Principal Investigator will obtain the required data from all participating sites and will report to the SMC annually, or as otherwise directed by the SMC.

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18 Appendices Appendix A: Study Agent Background Information

This appendix contains background information about the study agents. This was provided by CTEP as a summary of relevant data for each study agent. More detailed information about each agent can be found in their respective investigational brochure.

18.1.1 CTEP supplied agents

18.1.1.1 MK-2206

The PI3K/AKT pathway is downstream of the common growth factor tyrosine kinase receptors (RTK), including EGFR, HER2, IGFR, etc., and is a likely driver of tumor progression in most carcinomas (127-129). AKT protein kinase is activated in a substantial proportion of human solid tumors (breast, endometrial, ovarian, prostate, pancreatic, colon, gastric, and non-small cell lung cancer [NSCLC]). Upregulation of AKT can be caused by direct amplification/mutation of AKT, or by overexpression of RTKs, PI3K and RAS, and/or by inactivation of the tumor suppressor PTEN. Because of its key function in cell survival, AKT plays a pivotal role in rendering tumor cells insensitive or resistant to chemotherapy or targeted agents.

The rationale for the use of an AKT inhibitor in treatment of various malignancies is included in the following references (3, 127-159). MK-2206 is the first allosteric inhibitor of AKT to enter clinical development (3). MK-2206 demonstrated AKT inhibition and antiproliferative activity as single agent and in combination with other agents in multiple human cancer cell lines, such as breast, ovarian, lung, and prostate. MK-2006 synergized antitumor effects of docetaxel, erlotinib, and carboplatin in vivo in various human tumor xenograft models.

Mechanism of Action

MK-2206 is a selective allosteric inhibitor of AKT (3). MK-2206 does not bind to the active site of AKT, and consequently does not compete with either ATP or peptide substrate for binding to AKT. It is equally potent against the two human AKT isoforms, AKT1 and AKT2, and ~5-fold less potent against AKT3.

Nonclinical Studies

In Vitro Activity Studies

In vitro single-agent activity of MK-2206

In an in vitro kinase assay with GSK3 alpha peptide as substrate, MK-2206 strongly inhibited kinase activity of the three human isoforms of AKT, AKT1, 2, and 3, with 50% inhibitory concentration (IC50) values of 8, 12, and 65 nM, respectively. MK-2206 exhibited no inhibition (IC50>50 micromol/L (mcM) against the (pleckstrin-homology domain)-deletion mutants of AKT, indicating that this domain is essential for binding MK-2206 to AKT. Apart from AKT, MK-2206 was tested at a single concentration of 1 mcM against a panel of ~250 proteins including kinases without demonstrating significant (≥50%) inhibition against any kinase.

The antiproliferative potency of MK-2206 was evaluated against a panel of tumor cell lines using in vitro proliferation and viability assays. Among 52 cell lines, 18 cell lines were highly

sensitive (IC50<1 mcM), 7 were moderately sensitive (IC50=1-5 mcM), and 27 cell lines were insensitive (IC50>5 mcM) to MK-2206. Highly sensitive cell lines included breast, ovarian, prostate, NSCLC, small cell lung cancer (SCLC), gastric, and endometrial cancer. At least one of the following genetic defects was represented in the majority of sensitive cell lines: PTEN mutation, PI3KCA mutation, AKT amplification, or genomic amplification of HER2 or MET. Nine of 27 cell lines that were insensitive to MK-2206 had either PTEN or PI3KCA mutation, or were deficient in PTEN protein. Among eight cell lines carrying RAS or BRAF activating mutation, six were insensitive to MK-2206.

MK-2206 activity in combination with other agents

In a proliferation/viability assay, various degrees of synergism between MK-2206 and lapatinib (a dual EGFR/HER2 inhibitor) were observed in eight breast cancer cell lines. MK-2206 and docetaxel demonstrated additive to synergistic antiproliferative activity against nine breast-cancer cell lines, however, the effects were dependent on agent sequence. Enhancement of antitumor activity occurred only when cells were treated first with docetaxel and then exposed to MK-2206. In contrast, co-administration of the two agents resulted in antagonism. Combination of MK-2206 with erlotinib produced various degrees of synergism in nine NSCLC cell lines, including A431 epidermoid cells overexpressing EGFR.

MK-2206 synergized antitumor activity of carboplatin, gemcitabine, doxorubicin, camptothecin, or 5-FU in ovarian, prostate and NSCLC cell lines. Antitumor activity of MK-2206/carboplatin, demonstrated in the ovarian cell line A2780, was accompanied with enhanced apoptosis (cleavage of caspases 3 and 7). However, the effect was dependent on the sequence of agent administration, as it occurred only when cells were exposed to carboplatin before or simultaneously with MK-2206. Enhancement of apoptosis did not occur when MK-2206 preceded carboplatin.

In Vivo Activity Studies

MK-2206 monotherapy administered as a single dose (10-240 mg/kg) to mice bearing human ovarian tumor (A2780) potently inhibited phosphorylation of AKT1/2 in blood as well as in tumor tissue. After a single dose of 30, 120, or 240 mg/kg of MK-2206, ≥80% inhibition of AKT1/2 was achieved in peripheral blood; inhibition persisted at this level for 6 and 24 hours at the 120- and 240-mg/kg dose, respectively. Pronounced inhibition (70-90%) of phosphorylation of AKT1/2 (pAKT1/2), lasting for at least 24 hours, was detected also in tumor tissue following the single 120- or 240- mg/kg dose. MK-2206 combinations with docetaxel, carboplatin, or erlotinib exhibited significantly more potent antitumor activity than each agent in monotherapy settings. For example, a combination of MK-2206 and docetaxel, administered for 4 weeks as one weekly intravenous (IV) dose of docetaxel (30 mg/kg) followed 24 hours later by MK-2206 (240 or 480 mg/kg) given orally (PO) once a week (QW), produced strong antitumor responses in the A2780 ovarian-cancer xenograft mouse model. MK-2206 monotherapy was ineffective at all doses. Although the combination was significantly more effective than docetaxel monotherapy, responses did not endure beyond the treatment period. Tumor regression was also observed in PC-3 prostate xenograft model following sequential treatment with docetaxel and MK-2206 (24 hours later). MK-2206 (120 mg/kg) was given three times per week (days 1, 3, 5) and docetaxel (5 mg/kg) once weekly on day 0. Inhibition of tumor growth was also enhanced

when MK-2206 was combined with carboplatin and erlotinib in the NSCLC H460 and H292 xenograft models, respectively. Overall, the agents' combinations were well-tolerated at the effective dose levels, although transient 10-20%-weight loss was observed in some animals.

Pharmacokinetics

Pharmacokinetic (PK) parameters of MK-2206 in the rat, dog, and monkey are summarized in Table 18. Across the species, MK-2206 showed moderate plasma clearance (Cl) and high volume of distribution at steady state (Vdss) and elimination half-life (t1/2) ranging from 4 hours in rats to >12 hours in dogs and monkeys.

Table 18. Summary of nonclinical pharmacokinetics for MK-2206

IV Route								PO Route	
Species	Dose (mpk)	Cl (mL/min/kg)	Vdss (L/kg)	t1/2 (h)	Dose (mpk)	AUC (mcM·h)	Cmax (mcM)	tmax (h)	F (%)
Rat	2	27.6	9	4.2	10	2.1	0.29	5	20
					100	35.2	2.76	2.7	35
Dog	0.5	7.7	8	12.5	2	8.89	0.38	4	83
Rhesus	0.5	10.8	11	14					

Cl: clearance; Vdss: volume of distribution at steady state; t1/2: elimination half-life; AUC: the area under the time-concentration curve; Cmax: maximum drug concentration in plasma; tmax: time needed to achieve Cmax; F: oral availability; mcM: micromol/L; mpk: mg per kg;

Vehicle: intravenous (IV): DMSO, oral route (PO): 0.5% methylcellulose (10 mpk), 30% Captisol (100 mpk), or 1% methylcellulose (3 mpk)

MK-2206 was significantly bound to plasma proteins (ranged from 96% to 88% in mice>rats>dogs>monkeys>humans) with moderate Cl in rats, but relatively low Cl in dogs and monkeys.

The oral availability of MK-2206 was acceptable in rats (20%-35%) and better in dogs (83%). Plasma Cl in rats occurred primarily by direct glucuronidation with <3% of MK-2206 excreted in feces. In dogs, elimination occurred via multiple metabolic pathways including oxidation followed by glucuronidation, direct glucuronidation, and formation of a carbamoyl glucuronide. Relative to rats, a higher fraction (30%) of parent compound MK-2206 was excreted (urine, bile, and feces) in dogs. Significant intestinal secretion was observed in dogs. No metabolites of MK-2206 with biologic activity were noted.

MK-2206 was also metabolized by oxidation in human liver microsomes, primarily via the 3A4 isoenzyme of the P450 cytochrome (CYP) enzyme complex. MK-2206 was neither a potent inhibitor nor inducer of human CYPs. Transport experiments in the P-glycoprotein (P-gp)-transfected cell lines (L-MDR1) suggested that MK-2206 could be a P-gp substrate. In addition, MK-2206 demonstrated weak inhibition of vectorial transport of digoxin in L-MDR1 cells (IC50 of 13.4 mcM).

Toxicology

In the 10-day tolerability studies, dose-limiting toxicities (DLTs) were observed at the dose of ≥200 mg/kg/day and ≥15 mg/kg/day in rats and dogs, respectively. In a 4-week safety study in dogs, MK-2206 was administered at 2.5, 5, or 10 mg/kg PO every-other-day (QOD) followed by a 2-week recovery period. The 10-mg/kg dose was poorly tolerated as manifested by severe body-weight loss and other physical and histomorphological signs of toxicity, which required cessation of dosing by the end of the second week of treatment. The 5-mg/kg dose was tolerated and although treatment-related toxicities occurred, they were transient. As no significant toxicities were observed in dogs at the 2.5-mg/kg dose, this dose was defined as NOAEL (no observed adverse effect level). Exposure at this level characterized by the maximum plasma concentration (C_{max}) and the area under the time-concentration curve (AUC_{0-48h}) corresponded to 0.37 mcM and 8.52 mcM*hr, respectively. A potential safety concern associated with MK-2206 therapy is prolongation of the QT-corrected (QTc) interval, which seemed to be dose-dependent. While QTc-prolongation was persistent in dogs at the 10-mg/kg dose, it declined and returned to baseline between 24 to 72 hours post-dosing at 5 and 2.5 mg/kg. No cardiovascular changes were observed at the 1-mg/kg dose of MK-2206 (C_{max} of 0.092 mcM and AUC_{0-24h} of 1.6 mcM) within 48 hours following dosing in dogs.

A tissue distribution study demonstrated that [14C]MK-2206 was widely distributed in Sprague Dawley (albino) and Long Evans (pigmented) rats except for the central nervous system. The majority of the radioactivity went into the muscle, liver and skin shortly after dosing. In Sprague Dawley and Long Evans rats, the radioactivity in most tissues was comparable. It declined in parallel to that in blood and became negligible 3 days post-dose. However, the radioactivity was more sustained at higher levels in the skin and the uveal tract of the eye in Long Evans than in Sprague Dawley rats. The concentration (ng equivalent [14C]MK-2206/g tissue) in the skin and the uveal tract of Long Evans rats was 37- and 84-fold higher than that in the respective tissues of Sprague Dawley rats 24 hours post-dose.

An additional safety concern involves MK-2206-induced hyperglycemia and hyperinsulinemia. MK-2206 induced hyperglycemia in all preclinical species tested. In the most sensitive species, the dog, the glucose level was elevated by 24%-35%, when MK-2206 was administered at 5 mg/kg QOD for 4 weeks (AUC0-48 of 19.6 mcM·h and Cmax of 0.73 mcM). Such exposure is expected to correspond with human exposures achievable at the upper MK-2206 dosing range.

Results from genetic toxicology assays demonstrated that MK-2206 was neither genotoxic nor mutagenic.

Clinical Development of MK-2206

Preliminary clinical PK/pharmacodynamic and safety experience is derived from a Merck (Merck Sharp and Dohme)-sponsored phase 1 study in healthy volunteers (HVs) and company phase 1 studies in patients with advanced solid tumors.

Projections derived from preclinical PK and metabolism studies in dogs suggested that the target exposure corresponding to a plasma concentration of ≥100 nM MK-2206 over 8 hours and AUC0-48h of ~2 mcM·h in humans could be attained by MK-2206 dosed at 30-70 mg QOD on a 28-day cycle schedule. Clinical PK/pharmacodynamic data confirmed that the MK-2206 dose of 60 mg QOD conferred substantial and lasting inhibition of AKT as measured in tumor biopsies from cancer patients. The 60-mg QOD dose level is being currently investigated as the maximum tolerated dose (MTD) in the expanded cohort of patients.

Preclinical and clinical experience from Merck-sponsored studies suggests potential benefit of a less frequent dosing schedule (i.e., QW). It is potentially feasible to administer MK-2206 at higher dose levels on a less frequent dosing schedule to maximize significant or peak target inhibition. This approach may also alleviate DLTs (e.g., skin rash) associated with accumulated exposure to MK-2206. Dose escalation on a QW schedule is currently being evaluated at the MK-2206 doses ranging from 90-300 mg.

Clinical Pharmacokinetics

The clinical PK for MK-2206 was evaluated in HVs receiving a single dose (0.25-100 mg) and in cancer patients given either a 30-, 45-, 60-, 75-, or 90-mg dose on the QOD schedule or 90-, 135-, 200-, 250-, or 300-mg dose on the QW schedule.

Although Day 1 $AUC_{0\text{-}48h}$ and C_{max} values achieved at ≤90 mg in cancer patients overlapped with the ranges observed in HVs, overall, MK-2206 exposures in cancer patients trended on average somewhat higher than those observed in HVs. In all cohorts evaluated at ≤200 mg QW, exposures after the first and last dose in Cycle 1 were below the dog NOAEL $AUC_{0\text{-}48h}$ and C_{max} values of 8.52 mcM*hr and 365 nM, respectively. The mean first dose MK-2206 $AUC_{0\text{-}48h}$ and C_{max} were 1.77 mcM*hr and 62.2 nM, respectively, for 60 mg QOD, and 14.8 mcM*hr and 466 nM, respectively, for 300 mg QW. The dog NOAEL exposures were exceeded in humans following the first dose of 300 mg QW.

The variability in $AUC_{0\text{-}48h}$ and C_{max} following the first dose, where it could be assessed, was low to moderate across all dose levels, with the coefficient of variation (CV) values ranging from approximately 10%–60%. From a visual inspection of the data there does not appear to be a substantial or consistent departure from dose proportionality for either $AUC_{0\text{-}48hr}$ or C_{max} following the first dose up to the 300-mg dose level.T_{max} and apparent terminal $t_{1/2}$ values from cancer patients were generally within the ranges observed in HVs. Median T_{max} values ranged from 4–10 hours across all dose regimens, and harmonic mean apparent terminal half-life ($t_{1/2}$) values ranged from approximately 60–80 hours, with the exception of the 90 mg QOD cohort. At 90 mg QOD, apparent terminal $t_{1/2}$ was assessable in one patient and was approximately 50 hours.

Clinical Efficacy/Pharmacodynamics

Preliminary pharmacodynamic results in cancer patients indicate that phosphorylation of AKT in whole blood is substantially inhibited at all dose levels evaluated on the QOD and QW schedule.

Additionally, preliminary results indicate that substantial pAKT inhibition was demonstrated in tumor tissue at 60 mg QOD. As there is a causal relationship between the development of hyperglycemia/hyperinsulinemia and mechanism of AKT inhibition, such events could potentially implicate pharmacodynamic activity of MK-2206. In cancer patients, reversible grade 1/2 hyperglycemia was observed across all dose levels in a total of 59 patients. Hyperinsulinemia occurred in 26 patients who received MK-2206 60 mg QOD. From a preliminary analysis, these adverse events (AEs) do not appear to be dose-dependent. Neither hyperglycemia nor hyperinsulinemia was observed at any single dose in HVs.

Thus far, no formal efficacy studies have been performed with MK-2206; however, in patients with advanced solid tumors, early indications of antitumor activity included substantial decreases in CA125 in some patients with ovarian cancer and PSA stabilization in some prostate cancer patients. Minor RECIST responses, e.g., <30% decreases in tumor size, have also been observed in a patient with melanoma (16%), a patient with pancreatic cancer (23%), and in a patient with neuroendocrine tumor (20%). No partial responses, e.g., confirmed >30% decreases in tumor size, have been observed.

Clinical Toxicology

Preclinical efficacy and safety studies and preliminary safety data from the clinical studies support the use of MK-2206 via the oral route, both as monotherapy and in combination with other anticancer agents.

Overall, MK-2206 has been generally well-tolerated when administered as a single PO dose (0.25-100 mg) to HVs, or to cancer patients as the 30-60 mg PO dose on the QOD schedule and the 90-200 mg PO dose on the QW schedule. Mild to moderate skin rash was observed in 21 of 42 patients (50.0%) and severe skin rash was observed in 5 of 42 patients (11.9%) at the dose of 60 mg QOD. Skin rash resolved following the 1- to 2- week therapy break. The higher doses evaluated in oncology patients (i.e., 75 and 90 mg QOD and 300 mg QW) were not tolerated and resulted in DLT of grade 3/grade 4 skin rash.

Mild to moderate mucositis and conjunctivitis were associated with rash. The supportive-care measures included hydration, topical steroid preparations, oral corticosteroids, oral antihistamines, and oral antibiotics. Other common AEs included nausea, fatigue, vomiting, and diarrhea. These AEs were mild to moderate and in most cases were resolved by the standard supportive care and did not require therapy modifications. Hyperglycemia and hyperinsulinemia, both expected mechanism-based AEs, were observed in approximately 76% and 57% of patients, respectively, receiving MK-2206 on the QOD schedule. Episodes were generally mild, transient, and did not require therapeutic intervention. Importantly, administration of insulin may not counteract MK-2206-induced hyperglycemia due to mechanism-based insulin resistance. In this case, hydration and oral antihyperglycemic agents can be used as the supportive-care measures. Grade 3 hyperglycemia occurred in one patient who received 60 mg QOD and required treatment with PO antihyperglycemic medication. Grade 4 hyperglycemia was reported in one patient who received MK-2206 45 mg QOD in combination with erlotinib. In addition to supportive care measures, blood glucose management included administration of insulin and PO antihyperglycemic medication.

Grade 1/grade 2 prolongation of QTc-interval was observed in 14 out of 64 patients (21.9%) with available 12-lead ECG data. Prolongations ≥30 msec but <60 msec occurred in 4 of these patients. Grade 3 QTc prolongation was reported in one patient who received 135 mg QW. Episodes of QTc interval prolongation were in general isolated with no apparent dependency on dose or exposure levels, and were not considered clinically significant by investigators and were not reported as adverse experiences. Eleven patients experienced sinus bradycardia (<50 bpm) during Holter or ECG monitoring. These events were asymptomatic and were not clinically significant. While a causal relationship between these events and administration of MK-2206 is uncertain, similar side effects were seen preclinically in conscious dogs. Consequently, patients with a history or current evidence of heart disease should be excluded from enrollment on MK-2206 trials. Standard 12-lead ECG measurements should be performed at the protocol-specified time-points.

Developmental/Reproductive Toxicity

Developmental and reproductive toxicity studies of MK-2206 have not been performed thus far. MK-2206 was not tested in pregnant or breast-feeding women. Women of child-bearing potential and men participating in clinical studies of MK-2206 must use appropriate contraception, including abstinence and double-barrier methods, throughout MK-2206 therapy. In preclinical mutagenicity studies, MK-2206 was neither genotoxic or mutagenic.

Drug Interactions

No clinical drug interaction studies have been performed with MK-2206. Oxidative metabolism of MK-2206 in human liver microsomes was catalyzed primarily by CYP3A4. MK-2206 was not an inhibitor of the major CYPs. MK-2206 was shown to be a P-gp substrate.

Weekly dosing

Preclinical studies in animal models have shown evidence of improved efficacy of the once weekly (QW) dosing in mono-therapy using the A2780 xenograft (PTEN mutant) model in two independent studies. Additionally, QW dosing of MK-2206 results in similar or enhanced single agent and combination efficacy in NCI-H-292 Xenografts. In the phase I clinical trial PN002, MK-2206 showed a Cmax = 170 nM (after 1st dose) and Cmax = 225 nM (SS), which could potentially lead to better tumor penetration, more tumor cytotoxicity and better efficacy in comparison to the every other day (QOD) mode of administration. Mean Coverage above target PK was 133 hours (out of 168 hours), which may potentially alleviate skin toxicity. Furthermore, this dosing schedule is significantly more convenient for the patient. More detailed information and updates about the rationale for the use of QW dosing can be found in the investigational brochure.

16.1.1.2 AZD6244

AZD6244 (ARRY-142886) is a potent, selective, orally-available, and non-ATP competitive small molecule inhibitor of the mitogen-activated protein (MAP) kinase kinase, MEK-1/2 (Investigator's Brochure, 2009; Friday and Adjei, 2008). AZD6244 inhibited the activity of purified MEK enzyme with an IC50 of 10-14 nM, and was found to be inactive or only

minimally active at 10 □M against a panel of other kinases, including epidermal growth factor receptor (EGFR), ERB2, p38□, ERK2, and MKK 6 kinases. Because ERK is the only known substrate of MEK, the inhibition of MEK will target only the ERK signal transduction pathway and other signal transduction pathways will not be blocked. AZD6244 is metabolized to biologically active N-desmethyl AZD6244 which is more potent than the parent compound. In vitro, in vivo and preliminary results from clinical studies suggest that AZD6244 exhibits a favorable pharmacologic and toxicologic profile.

The RAS/RAF/MEK/ERK signaling pathway plays a central role in the regulation of many cellular processes including proliferation, survival, differentiation, apoptosis, motility, and metabolism (O'Neill and Kolch, 2004; Wellbrock et al., 2004). This pathway is one of the most important and best understood MAP kinase signal transduction pathways, activated by a diverse group of extracellular signals including integrins, growth factor receptors (i.e., EGFR, platelet-derived growth factor receptor [PDGFR], and insulin-like growth factor-1 receptor), and cytokines (Janssen et al., 2005). Activated RAS triggers the phosphorylation and activation of RAF kinase which then phosphorylates MEK1 and MEK2 on 2 serine residues (Ahn et al., 2001). Activated MEK phosphorylates its only known substrates, ERK1 and ERK2. Phosphorylated ERK dimerizes and translocates to the nucleus (Khokhlatchev et al., 1998) where it is involved in several important cellular functions, including cell proliferation.

Overexpression of growth factors or growth factor receptors involved in the RAS/RAF/MEK/ERK pathway and activating genetic mutations of the signaling proteins may lead to uncontrolled proliferation and tumor formation. For example, RAS genes are the most frequently mutated oncogenes detected in human tumors (Janssen et al., 2005). RAS proteins are guanine nucleotide binding proteins that activate RAF proteins when bound to GTP. Cancer-associated mutations in RAS proteins stabilize the GTP-bound form of RAS, thereby providing a constitutive signal downstream in the cascade. In addition to being found in almost all pancreatic adenocarcinomas, RAS mutations are found in ~50% of colorectal carcinomas, 25-50% of lung adenocarcinomas, and also in some breast or ovarian cancers. BRAF mutations have also been observed in many human cancers, particularly melanoma (30-60%), thyroid (30-50%), colorectal (5-20%), and ovarian (~30%) cancers (Wellbrock et al., 2004; Friday et al., 2008). These mutations in BRAF usually involve gain-of-function substitutions that render the kinases constitutively active. Also, studies of primary tumor samples and cell lines have shown constitutive activation or overactivation of the MAP kinase pathways in cancers of the pancreas, colon, lung, ovary, and kidney (Hoshino et al., 1999). Therefore, agents targeting the RAS/RAF/MEK/ERK pathway may inhibit oncogenic signaling in tumor cells.

Nonclinical Studies

Efficacy

In vitro studies have shown that AZD6244 is potent and selective inhibitors of MEK (Investigator's Brochure, 2009). However, significant biochemical activity was not detected when tested at 10 µM against a diverse panel of 305 other molecules, including enzymes, receptors, kinases, transporters, and ion channels. The effects of AZD6244 on ERK phosphorylation and cell viability were determined in a panel of cell lines in which the mutational status of RAF and RAS are known. AZD6244 inhibited ERK1 and ERK2

phosphorylation with IC50 ranging from 0.0018 to 0.041 □M. AZD6244 was particularly potent in inhibiting the viability of cell lines with V600E BRAF gene mutation and some cell lines with KRAS mutations. Sensitive cell lines included those derived from colorectal, non-small cell lung cancer (NSCLC), pancreatic, and melanoma tumors. Two metabolites of AZD6244 (N-desmethyl AZD6244 and an amide AZD6244) have been identified. The N-desmethyl metabolite was found to be 3-to 5-fold more active than the parent compound in cellular ERK phosphorylation inhibition assays, whereas the amide metabolite was up to 50-fold less active than AZD6244. In tumor cell viability inhibition assays, N-desmethyl metabolite was ≥5-fold more potent in inhibiting cell viability.

Significant suppression of tumor growth in response to AZD6244 treatment was observed in several xenograft mouse models derived from a range of tumor types including melanoma, breast, pancreatic, lung, colon, and hepatocellular carcinomas. (Tran et al., 2006; Breikreutz et al., 2007; Davies et al., 2007; Tai et al., 2007). In papillary thyroid cancer models, AZD6244 effectively inhibited tumor growth, both in vitro and in vivo, particularly in tumor cells carrying activating BRAF gene mutations (Ball et al., 2007). In the Calu-6 lung cancer xenograft model, AZD6244 suppressed tumor growth at doses of 10, 25, or 100 mg/kg given twice daily (BID), and the minimal effective dose was identified as 0.75 mg/kg administered BID (Davies et al., 2007). In this model, MEK activity was inhibited as assessed by determination of phosphorylated ERK (pERK) levels in tumor. Studies using human colorectal xenograft models demonstrated that AZD6244 inhibited tumor growth by inhibition of cell proliferation in SW620 model and by induction of apoptosis in Colo205 model. In vivo studies of the KRAS mutation-positive (KRAS+) human cancer xenografts have demonstrated potential for additive /synergistic effects of AZD6244 in combination with a number of cytotoxic and targeted agents, including docetaxel, irinotecan, gemcitabine, Iressa (gefitinib), AZD2171 (cediranib), rapamycin, cisplatin, and temozolomide (Investigator's Brochure, 2009).

Pharmacokinetic/Pharmacodynamic Studies

Two oral formulations of AZD6244 have been tested in preclinical pharmacology studies: the original mix-and-drink formulation (AZD6244 free-base), and the AZD6244 hydrogen sulfate salt (capsule) formulation (AZD6244 Hyd-Sulfate) (Investigator's Brochure, 2009). The former requires reconstitution of the AZD6244 free-base crystalline powder in the 25% (w/v) Aqueous Captisol®(sulfobutyl ether β-cyclodextrin) (SBE-CD) solution. Absorption of AZD6244 free-base was moderate to high at low doses in rat, dog and monkey, but showed decreasing bioavailability with increasing dose. The declining bioavailability at higher doses may be due to the low aqueous solubility of AZD6244. When administering AZD6244 hydrogen sulfate, AZD6244 exposure, expressed as an area under the plasma concentration-time curve from 0 to 12 hours (AUC0-12), increased roughly in proportion with dose in the mouse and monkey. In 1-month studies, 3 to 6-fold greater exposures were achieved with AZD6244 hydrogen sulfate at 7.5 mg/kg BID than with the AZD6244 free-base at 30 mg/kg BID. There was no/minimal accumulation of AZD6244, whether dosed with AZD6244 free-base or AZD6244 hydrogen sulfate, on multiple dosing in the mouse, rat, or monkey. Oral bioavailability of AZD6244 in monkey, following dosing with 5 mg/kg of either free-base or capsule, was 29% and 56%, respectively. N-desmethyl metabolite was not detectable in rat and at only trace levels in the monkey, but was produced in mouse at circulating levels around 2-12% of parent compound.

Studies in rats indicate that AZD6244 is widely distributed, although tissue concentrations were lower than blood concentrations. High levels of protein binding (93.7-99.7%) were observed in all preclinical species tested and in humans (98.4%). There was no evidence of AZD6244-related material binding to melanin and minimal penetration into the central nervous system (CNS). □ AZD6244 was metabolized by cytochrome P450 (CYP enzymes) 1A2, 2C19 and 3A4, with CYP1A2 being the enzyme primarily responsible for the formation of the N-desmethyl metabolite. Glucuronidation appears to be a significant clearance mechanism for AZD6244 and N-desmethyl metabolite. AZD6244 did not inhibit CYP 1A2, 2C8, 2C19, 2D6, and 3A4. It was a weak inhibitor of CYP2C9 (IC50 44.7 µM). N-desmethyl AZD6244 did not inhibit CYP 2A6, 2C8, 2C9, 2C19, 2D6, 2E1, or 3A4, but was a weak inhibitor of 1A2 (IC50 of 18.9 µM). In rat, mouse and monkey, fecal excretion was the predominant route after oral and intravenous dosing.

Toxicologic Studies

The toxicologic effects of AZD6244 were evaluated in acute dose (single or two doses on a single day) and 1-month, repeat-dose studies in Sprague-Dawley rats and cynomolgus monkeys (Investigator's Brochure, 2009). The repeat-dose study in rats indicated that the agent was well tolerated but produced soft stools and gastrointestinal mucosal mineralization associated with increased serum phosphorus and decreased albumin. Diarrhea, dehydration, electrolyte imbalance, and secondary renal changes were observed in monkeys. The vehicle, Captisol®, used to reconstitute AZD6244 free-base powder, is known to cause gastrointestinal disturbances in preclinical studies. A reduction in the volume of Captisol® administration to monkeys was associated with a reduced incidence of diarrhea; however, it is likely that AZD6244 exacerbated the vehicle effects. The no observable adverse effect levels (NOAELs) in rats (female) and monkeys in 1-month studies were identified at 10 mg/kg/day BID; a NOAEL was not achieved in male rats. Dosing with AZD6244 hydrogen sulfate (0.5, 1.5, and 4 mg/kg BID) for up to 6 months to monkeys was also associated with fluid and/or red-colored feces, but with no notable gastrointestinal tract or renal pathology. Tissue mineralization was not apparent in cynomolgus monkeys dosed for up to 6 months with AZD6244 hydrogen sulfate. However, mineralization was seen in multiple tissues (cornea, kidney, liver, myocardium, skeletal muscle, glandular stomach) in mice dosed with AZD6244 hydrogen sulfate for up to 1 month, and in the livers of a small number of mice dosed up to 6 months.

AZD6244 and its N-desmethyl metabolite showed no evidence of mutagenic potential, but AZD6244 produced an increase in micronucleated immature erythrocytes in mice, predominantly via an aneugenic mode of action. AZD6244 showed enhanced cytotoxicity in the presence of ultraviolet (UV) light. With the exception of tissue mineralization in rats and mice, there was evidence of reversibility of most changes in the 1- or 6-month studies with AZD6244 (free base or hydrogen sulfate). Reproductive toxicology data in mice indicate that AZD6244 can have adverse effects on embryofetal development and survival at dose levels that were not toxic to mothers.

In summary, preclinical studies demonstrated that AZD6244 exposures can be significantly enhanced by using AZD6244 hydrogen sulfate, compared to AZD6244 free-base. In 6-month toxicology studies, AZD6244 exposures (AUC 0-12) achieved with AZD6244 hydrogen sulfate at high dose levels (4 mg/kg BID in monkeys and 20 mg/kg BID in mice) were approximately 3-

fold and 15-fold higher in primates and mice, respectively, when compared with those achieved in man at 75 mg BID AZD6244 hydrogen sulfate. At the NOAEL of 1.5 mg/kg BID from 1-month and 6-month studies in primates, exposures were generally similar to those seen in man after dosing at 75 mg BID AZD6244 hydrogen sulfate. Exposure achieved in the 6-month mouse study at the low dose level of 1 mg/kg BID was slightly below that seen to date in man after dosing with AZD6244 hydrogen sulfate (75 mg BID). Preclinical pharmacology studies suggest an acceptable safety profile for the administration of AZD6244 free base or AZD6244 hydrogen sulfate to human cancer patients.

Clinical Experience

AstraZeneca sponsored a phase 1 open-label study designed to assess the safety, tolerability and pharmacokinetics (PK) of AZD6244 hydrogen sulfatein patients with advanced solid malignancies. In addition, this study planned to assess the relative bioavailability of the capsule vs. free-base formulation. This is the first clinical study that employed the AZD6244 capsule formulation. The further development of AZD6244 is to be continued with the hydrogen sulfate salt (capsule formulation) (Investigator's Brochure, 2009).

This study was conducted in two parts. Part A (31 patients enrolled) of the study was a dose escalation study, was designed to provide adequate tolerability, safety, PK, and pharmacodynamic data. In Part A, the first cohort received a single 25 mg dose of the AZD6244 hydrogen sulfate on day 1, followed by BID dosing from day 2 onwards. Other doses investigated were 50 mg, 75 mg, and 100 mg. The aim of Part B (29 patients randomized) was to determine the relative oral bioavailability of the AZD6244 hydrogen sulfate, and secondly to expand the safety, tolerability and preliminary efficacy data for the maximum tolerated dose (MTD) of 75 mg BID. In Part B, patients received either a single dose of capsule formulation or free-base suspension formulation on day 1 and 8, followed by continuous BID dosing of capsule formulation from day 9 onwards.

An AstraZeneca-sponsored multi-arm combination study of the AZD6244 capsule formulation that evaluates PK interactions of AZD6244 with selected chemotherapy agents (docetaxel, dacarbazine, erlotinib, temsirolimus) is ongoing.

Pharmacokinetics

AZD6244 plasma PK parameters of AZD6244 hydrogen sulfate were similar after single and multiple dosing, suggesting the minimal accumulation over time after BID dosing. The AZD6244 exposure parameters, i.e., the maximum plasma concentration (Cmax) and the area under time-concentration curve (AUC) were approximately dose proportional across the 25- to 100-mg BID dose range after single (day 1) or multiple dosing studied on days 1, 8, 15, and 22: the geometric mean values of exposure parameters were: Cmax=369-1489 ng/mL and AUC=1361-7055 ng·h/mL on day 1, and Cmax=458-1365 ng/mL and AUC0-12=1515-4758 ng·h/mL on day 8. AZD6244 was absorbed relatively quickly across all dose levels, with a median time-to-reach Cmax (tmax) of 1.5 hours. Following the peak, AZD6244 concentration declined multi-exponentially with a mean terminal elimination half-life (t1/2) ranging from 5 to 7 hours, which was consistent across dose levels. The steady state volume of distribution (Vss) and clearance (Cl) also remained largely consistent across the dose range, with mean values

ranging from 87 to 126 L and 12 to 19 L/h, respectively. Plasma concentrations of N-desmethyl AZD6244 followed a similar PK profile as that of AZD6244, although exposure was much lower, achieving Cmax and AUC values generally <15% of the parent compound, in each patient. The median tmax was approximately 1.5 hours and t1/2 ranged from 9-13 hours. The AZD6244 amide metabolite showed increased exposure on multiple dosing indicating some accumulation. Concentrations of the AZD6244 amide varied greatly among patients and t1/2 could not be assessed. Given the 3-to 5-fold greater potency of the N-desmethyl metabolite compared to AZD6244 shown by the in vitro cell-based ERK phosphorylation assay, the former is likely to contribute to pharmacodynamic effects. In contrast, AZD6244 amide, which was approximately 40- to 50-fold less active than AZD6244 in this assay, is unlikely to contribute significantly to AZD6244 biological activity. Nevertheless, both metabolites will be measured in all future AstraZeneca-sponsored clinical studies of AZD6244 hydrogen sulfate.

The capsule formulation was demonstrated to significantly improve the oral bioavailability, although large inter-patient variability was noted. The estimated oral bioavailability of capsule relative to the free-base suspension based on a dose-normalized AUC0-24 was 263% (90% CI = 214 to 322%). The geometric mean values of the exposure parameters obtained for AZD6244 hydrogen sulfate at 75 mg BID (MTD) were: AUC0-24 = 6335 and 5448 ng·h/mL on day 1 and 8, respectively, and Cmax = 1207 and 1439 ng/mL on day 1 and 8, respectively). In comparison, the exposure achieved by hydrogen sulfate at the MTD was significantly higher than that achieved by the free base suspension at 100 mg BID (MTD); the estimated exposures by hydrogen sulfate relative to those by free-base, achieved at their respective MTDs, were 197% (90% CI = 161 to 242%) and 252% (90% CI = 182 to 348%) based on AUC0–24 and Cmax, respectively.

A food effect study involving administration of AZD6244 hydrogen sulfate to patients with advanced solid malignancies under fasting conditions and with a high-fat meal indicated a statistically significant effect of food on the exposure of AZD6244. Geometric mean Cmax and AUC values were reduced by approximately 60% and 20%, respectively, under fed conditions. Therefore, it is recommended for further clinical studies that AZD6244 should continue to be taken on an empty stomach (no food or drink other than water for 1 hour prior to dosing and 2 hours after dosing).

There is no evidence of a PK interaction between AZD6244 and either docetaxel or dacarbazine, when given in combination as either twice daily dosing of AZD6244 50 mg or 75 mg with docetaxel 75 mg/m2 on day 1 of a 21 day cycle, or as twice daily dosing of AZD6244 50 mg or 75 mg with dacarbazine 1000 mg/m2 on day 1 of a 21 day cycle.

Pharmacodynamics

Inhibition of phosphorylation of ERK as an indirect measure of MEK inhibition by AZD6244 hydrogen sulfate was assessed in PBMCs from each patient at 4 time-points post-single-dose administration, across the dose levels (25-100 mg). Inhibition of ERK phosphorylation was observed at all doses, with the greatest inhibition generally occurring at the first time point (1-hour post dose); 73% inhibition was seen at 1-hour at the 75-mg BID dose, which declined to ~23% at 8-hour time-point.

Clinical Activity

In the AZD6244 hydrogen sulfate single-agent study, 55 patients had RECIST evaluable tumors. There was one complete response in the 75-mg dose cohort in Part A after 16 weeks. This patient had a BRAF+ melanoma. At the 75-mg dose, 16/35 (45.7%) patients had stable disease for ≥6 weeks. Nine patients in Part A and 13 patients in Part B had a best response of stable disease. Ten out of 55 (18.2%) patients (not including the patient who had a complete response) had stable disease of ≥16 weeks.

Safety

Twenty-eight patients were dosed in Part A of the study: 25 mg BID (n=6), 50 mg BID (n=7), 75 mg BID (n=7) and 100 mg BID (n=8). One dose-limiting toxicity (DLT) (grade 3 fatigue that resolved on discontinuation of AZD6244 hydrogen sulfate) was reported in the first 22 days of dosing in the 75 mg BID cohort. Two DLTs were reported in the first 22 days of dosing in the 100 mg BID cohort: grade 3 pleural effusion and grade 3 rash.

Based on the data from this study, the emerging safety and tolerability profiles for the AZD6244 capsule formulation are broadly consistent with both the patient population, and the adverse event profile seen with the AZD6244 free-base suspension formulation. The most frequently reported adverse events (AEs), regardless of dose cohort, severity, causality, or seriousness, were fatigue, acneiform dermatitis, nausea, diarrhea, and peripheral edema. All AEs of acneiform dermatitis were considered treatment related in all dose cohorts. In patients receiving 75 mg BID, all fatigue events were considered treatment related. Twenty-one patients reported at least one serious AE. Seven patients had at least one serious, treatment-related AE, with only hypertension being reported on more than one occasion. Thirty-three of 56 patients (58.9%) reported at least one grade 3 AEs during the study, of whom 18/56 (32.1%) experienced at least one treatment-related AE of ≥grade 3. Eighteen patients had permanent discontinuation of study treatment due to at least one AE (fatigue, anorexia, vomiting, nausea, left ventricular (LV) dysfunction, visual disturbance, fungal urinary tract infection, and acute renal failure). None of the 13 deaths occurred in this study was attributed to the therapy agent. Comparison of the AE frequencies from the AZD6244 hydrogen sulfate study and phase 2 monotherapy studies of AZD6244 free-base shows higher proportion of patients reporting the most frequent AEs, such as fatigue, dermatitis acneiform, diarrhea, nausea, and peripheral edema, with the capsule formulation. This may be due to the higher plasma exposures achieved with the capsule formulation, but may also be in part a consequence of more heavily pretreated patient population in the single-agent AZD6244 hydrogen sulfate (capsule) study. A higher proportion of patients withdrew due to AE at the MTD level (75 mg BID in the AZD6244 hydrogen sulfate study than at the AZD6244 free-base MTD (100 mg BID), although the different patient populations may be a contributing factor.

Vital signs

Small increases in mean systolic and diastolic blood pressure (SBP and DBP) were observed across most dose cohorts in Part A within the first week of treatment, although no significant dose-response relationship could be identified. In Part B a small mean increase in DBP was

observed after 1 week of BID dosing, reaching a maximum increase after 3 weeks BID dosing and being resolved by Week 12.

There were no clinically significant trends observed for pulse rate or percentage of oxygen saturation in blood (SpO2) at any dose in study. Mandatory assessment of LV dysfunction and ejection fraction (LVEF) was performed at baseline, after 8 weeks on study, and ad hoc on the occurrence of cardiorespiratory AEs. Data indicate a trend towards a mean decrease in LVEF at Week 8 across all dose cohorts, with no obvious dose-response relationship.

Review of the electrocardiogram (ECG) parameters demonstrated no consistent trend of significant prolongation of the corrected QT interval (QTc) across the dose cohorts in Part A, the maximum increase in mean QTc was noted in the 75-mg BID cohort with a 12.9 msec increase (standard deviation 13.2 msec, range –1 to +27 msec) 2 hours post-dose on day 8, n=5.

Laboratory parameters

There were a few minor differences in the clinical laboratory profile (primarily in renal and hematological parameters) between the AZD6244 free-base and hydrogen sulfate formulations; however, the information for hydrogen sulfate is based on a limited data set.

The AZD6244 capsule formulation contains vitamin E as an excipient. High doses of vitamin E have been reported to potentiate the anticoagulant activity of coumarins, such as warfarin. Therefore, patients who take such anticoagulants and receive the capsule formulation of AZD6244 will be advised to have an increased frequency of anticoagulation assessments, such as international normalized ratio (INR) measurements, upon starting AZD6244 therapy. In addition, the concomitant intake of excessive doses of vitamin E should be avoided in patients receiving the capsule formulation.

Based on preclinical reproductive toxicology studies, AZD6244 hydrogen sulfate (capsule formulation) should not be administered to pregnant or breast-feeding women, and conception while on treatment must be avoided.

Potential Drug Interactions

In vitro metabolic studies of AZD6244 using hepatocytes from humans and animals found that the biologically active N-desmethyl derivative was detected in mouse and human hepatocytes, was detected minimally in monkeys; and was not detected in rats (Investigator's Brochure, 2009). CYP1A2 was the enzyme primarily responsible for the formation of the N-desmethyl derivative; CYP2C19 and CYP3A4 were also minimally involved in the transformation. Neither AZD6244 nor its active metabolite were found to be inhibitors of CYP isoforms 1A2, 2C8, 2C19, 2D6, or 3A4. However, AZD6244 was found to be a weak inhibitor of CYP2C9 (IC50 = 44.7 uM) and the N-desmethyl derivative was a weak inhibitor of CYP1A2 (IC50 = 18.9 uM). Thus, at the systemic AZD6244 concentrations observed following 100 mg AZD6244 free-base formulation in man, no significant cytochrome P450 interactions would be expected. Since the formation of N-desmethyl AZD6244 from AZD6244 may occur through the CYP 1A2 pathway and smoking induces this pathway, the smoking status of the subjects should to be recorded in all studies (i.e., smoker or non-smoker) to investigate whether smoking status influences systemic drug exposures of N-desmethyl AZD6244.

18.1.1.3 Erlotinib

OSI-774 (erlotinib, Tarceva®) is an orally active, potent, selective inhibitor of the epidermal growth factor receptor (EGFR) tyrosine kinase (160). OSI-774 inhibits the human EGFR tyrosine kinase with a 50% inhibitory concentration (IC50) of 2 nM (0.786 ng/mL) in an in vitro enzyme assay and reduces EGFR autophosphorylation in intact tumor cells with an IC50 of 20 nM (7.86 ng/mL). OSI-774 inhibits EGF-dependent proliferation of cells at submicromolar concentrations and blocks cell-cycle progression in the G1 phase.

OSI-774 appears to bind specifically to EGFR. In a study of OSI-774 binding specificity, affinity of OSI-774 for 67 cellular receptors was examined (160). OSI-774 was shown to bind with low affinity to peripheral benzodiazepine (IC50=2.5 µM [980 ng/ml]), adenosine A1 (IC50=6.8 µM [2700 ng/ml]), and µ-opiate (IC50=7.0 µM [2800 ng/ml]) receptors. Binding affinities were 1250-fold higher than the IC50 concentration needed to inhibit purified EGFR tyrosine kinase (2 nM [0.79 ng/ml]). When tested at concentrations up to 1 µM (390 ng/ml), no significant inhibition of ligand binding to 64 other neurotransmitter receptors, regulatory binding sites, calcium channels, opioid receptors, or neurotransmitter uptake sites were observed.

Nonclinical Studies

In mice, daily oral administration of OSI-774 for 20 days inhibited subcutaneous (SC) growth of the HN5 human head and neck carcinoma in a dose-dependent manner as compared to vehicle-treated mice, with an estimated effective dose for 50% inhibition (ED50) of 9.2 mg/kg/day (160). Treatment with 11 mg/kg/day of OSI-774 immediately halted growth or slightly decreased the size of HN5 tumors >1 cm in diameter. The tumor stasis profile appeared to extend beyond the treatment period such that the tumor size for OSI-774-treated animals did not exceed pretreatment levels until at least 33 days post-treatment. Similar results were observed in A431 squamous cell carcinoma xenografts at a dose of 11 mg/kg/day OSI-774 over 20 days.

OSI-774 (10 mg/kg/day) has been studied in combination with cisplatin, doxorubicin, 5-fluorouracil, paclitaxel, vinorelbine tartrate, and gemcitabine (160). No antagonism of therapeutic efficacy was observed, and additive effects were observed with cisplatin (10 mg/kg intravenous [IV] daily × 1), doxorubicin (15 mg/kg IV daily × 1), paclitaxel (10 mg/kg intraperitoneal [IP] daily × 5), and gemcitabine (100 mg/kg IP three times daily × 4).

Nonclinical toxicology studies in rats and dogs have included acute and long-term general toxicology, genetic toxicology, reproductive toxicology, and local tissue tolerance of erlotinib. Clinical signs of toxicity in rats and/or dogs following a single dose of OSI-774 were dose dependent and included transient emesis, ataxia, papillary dilation, increased heart rate, decreased blood pressure, decreased activity, irregular respiration, convulsions, rapid chewing, salivation, and death. Following chronic administration in rats and dogs, the following toxicities were observed in at least one species: effects on the cornea (atrophy and ulceration); effects on the skin (follicular degeneration and inflammation, redness, and alopecia); atrophy of the ovary, lacrimal glands, and salivary glands; necrosis of the liver; papillary necrosis and tubular dilatation of the kidney; inflammation in the mandibular lymph nodes; hematopoiesis; delayed gastric emptying; and diarrhea. Increases in alanine aminotransferase, aspartate aminotransferase, and bilirubin were observed.

OSI-774 causes maternal toxicity with associated embryo-fetal lethality and abortion in rabbits at doses 3 times those in humans (AUC at 150 mg daily dose). However, when administered during organogenesis at plasma concentrations approximately equal to humans, no increase in embryo-fetal lethality or abortion in rats or rabbits was observed. Female rats treated with doses 0.3 to 0.7 times the human dose prior to mating and during the first week of gestation had an increase in early resorptions. No teratogenic effects were observed in rats or rabbits.

Pharmacokinetics

The total clearance of OSI-774 decreased with increasing dose, resulting in supraproportional increases in exposure (AUC) over the dose range of 1-2 mg/kg IV in rats and 0.5-7 mg/kg IV in dogs (160). In vitro, OSI-774 is slowly oxidized by liver microsomes. The majority of the absorbed dose is extensively metabolized in rats and dogs, and only a small amount is excreted as unchanged drug in urine, bile, and feces. The oral bioavailability of an aqueous suspension is 77% in rats and ~88% in dogs. Plasma protein binding of OSI-774 ranges from 92% to 95% in man, monkey, rat, and mouse, and is 85% in the dog. Corrected for protein binding of 95%, at the average plasma concentration responsible for 50% inhibition of tumor growth (oral dose of 9.2 mg/kg/day in the murine/human tumor xenograft model), the unbound concentration of drug in the plasma is estimated to be 86 nM (34 ng/mL). The estimated unbound concentration of OSI-774 in plasma is consistent (4-fold higher) with the IC50 for the in vitro cellular phosphotyrosine reduction assay and is 43-fold higher than the IC50 for the in vitro (isolated enzyme) tyrosine kinase assay. Finally, OSI-774 plasma protein binding depends on the levels of α-1-acid glycoprotein (AAG). Thus, AAG might be a significant determinant of pharmacokinetic and possibly pharmacokinetic–pharmacodynamic relationships in patients.

Pharmacokinetic Data

OSI-774 pharmacokinetics were examined in patients with advanced solid tumors who were treated daily for 21 days. Plasma samples were collected on Days 1, 3, and 21 for analysis. The following table lists the pharmacokinetic data for these patients:

Mean (%CV) OSI-774 Pharmacokinetic Parameters following Daily Dosing													
Dose (mg)	N	AUC0-24 (ng•h/ml)			R*		Cavg (ng/ml)			Cmax (ng/ml)			
		Day 1	Day 3	Day 21	Day 3	Day 21	Day 1	Day 3	Day 21	Day 1	Day 3	Day 21	
25	3	1580 (40)	2180 (49)		1.38 (11)		66	91		208 (38)	310 (40)		
50	6[a]	5650 (54)	5790 (57)	14900 (69)	1.59 (31)	1.83 (30)	235	241	621	508 (34)	716 (55)	682 (59)	
100	8[b]	9210	9080	32200	1.75	3.21	384	378	1340	765	1140	1740	

		(92)	(49)	(148)	(35)	(56)				(69)	(49)	(85)
150	7[c]	1420 0 (71)	2060 0 (22)	45800 (99)	1.74 (6.7	2.57 (47)	670	859	1910	1040 (70)	2040 (21)	1730 (82)
200	7	3600 0 (34)		74200 (29)		2.01 (42)	1360		3200	1420 (47)		2910 (29)

Abbreviations: AUC0-24, Area under the time-concentration curve from 0 to 24 hours; Cavg, daily average plasma concentration; Cmax, maximum serum/plasma concentration; CV, coefficient of variance; R, accumulation ratio

*Accumulation ratio = AUCDay 3 or 21/AUCDay 1

[a]Only 3 patients day 3, 3 subjects day 21

[b]Only 2 patients day 3, 3 subjects day 21

[c]Only 6 patients day 21

After oral ingestion, the OSI-774 Cmax values (Tmax) were achieved at a median of 3 hours (range, 2 to 12 hours). Both Cmax and AUC0-24 values were roughly proportional to the OSI-774 dose in the range of 25 to 200 mg/d (R2 = 0.33 and 0.46, respectively, on Day 1). Inter-subject variability was moderate at the 150-mg/d dose level, as indicated by coefficient of variation values of 64% for Day 1 AUC0-24 and Cmax.

Drug interactions

In *in vitro* human liver microsomes studies OSI-774 was slowly oxidized. OSI-774 appeared to be a substrate for CYP3A4, suggesting that OSI-774 could reduce the clearance of co-administered drugs whose metabolism is dependent on these P450 cytochrome isoenzymes.

- Erlotinib is both protein bound (92% to 95%) and metabolized by CYP3A4. Therefore, a potential for drug-drug interactions exists when erlotinib is co-administered with drugs that are highly protein-bound or CYP3A4 inhibitors or inducers.
- There is a potential interaction between erlotinib and warfarin. Patients have experienced elevated international normalized ratios (INRs) and bleeding with this combination of drugs. Patients on warfarin and erlotinib should have more frequent INR/prothrombin time (PT) determinations (e.g., weekly for the first month and weekly for a minimum of 2 weeks following Discontinuation of erlotinib).
- Proton pump inhibitor: Erlotinib's solubility decreases as the pH increases. Coadministration of omeprazole with erlotinib will decrease the AUC and Cmax by 46% and 61%, respectively.
- H2-antagonist: Avoid concomitant use of erlotinib with gastric acid-reducing agents if possible. When ranitidine 300 mg is given with erlotinib, erlotinib AUC and Cmax decrease by 33% and 54%, respectively. Increasing the dose of erlotinib will not compensate the loss of exposure. However, if an H2-antagonist receptor is needed, take

erlotinib at least 2 hours before or 10 hours following the H2-antagonist administration. Dosing such, erlotinib loss of exposure is minimized to AUC of 15% and Cmax of 17%.

- Smoking: Advise smokers to stop smoking while taking OSI-774. Smoking induces CYP1A2 enzymes and alters OSI-774 exposure by 64%.
- Food-drug interaction: Avoid grapefruit /grapefruit juice (potent CYP3A4) while taking OSI-774.

Safety profile

Adverse Events Associated with OSI-774

A comprehensive list of AEs possibly related to OSI-779 is provided in the CAEPR. Additional information can also be found in the Investigator's Brochure and the Package Insert. A few common or serious AEs are described below.

Rash or dermatosis: (Grades 1-3) has been reported in many subjects (~50%) during the first several days of treatment, although severity diminishes after 4 weeks of therapy. The use of topical agents (i.e., diphenhydramine, corticosteroids) and oral antibiotics (tetracycline) has been instituted in some patients with variable results. In patients with severe rash, treatment has been discontinued or the study drug dose reduced. The etiology of the rash is still unknown, but may be related to the mechanism of action of OSI-774.

Nausea and diarrhea: (Grades 1-2) have been observed in approximate half of all subjects treated with OSI-774. Both adverse events are transient and usually do not require a reduction in dosage. Diarrhea is well controlled in most patients by either the use of loperamide or dose reduction. All ongoing clinical trials recommend the institution of therapy with loperamide. As of June 2001, no significant drug interactions have been observed between OSI-774 and loperamide.

Hepatic toxicities: Cases of hepatic failure and hepatorenal syndrome (including fatalities) have been reported during use of OSI-779, particularly in patients with baseline hepatic impairment. Therefore, periodic liver function testing (transaminases, bilirubin, and alkaline phosphatase) is recommended. In the setting of worsening liver function tests, dose interruption and/or dose reduction with frequent liver function test monitoring should be considered. OSI-779 dosing should be interrupted or discontinued if total bilirubin is >3 x ULN and/or transaminases are >5 x ULN in the setting of normal pretreatment values.

Renal insufficiencies: Cases of hepatorenal syndrome, acute renal failure (including fatalities), and renal insufficiency have been reported. Some were secondary to baseline hepatic impairment while others were associated with severe dehydration due to diarrhea, vomiting, and/or anorexia, or concurrent chemotherapy use. In the event of dehydration, particularly in patients with contributing risk factors for renal failure (e.g., pre-existing renal disease, medical conditions or medications that may lead to renal disease, or other predisposing conditions including advanced age), OSI-779 therapy should be interrupted and appropriate measures should be taken to intensively rehydrate the patient. Periodic monitoring of renal function and serum electrolytes is recommended in patients at risk of dehydration.

Gastrointestinal perforation: Patients receiving OSI-779 are at increased risk of developing gastrointestinal perforation, which was observed uncommonly. Some cases had a fatal outcome. Patients receiving concomitant anti-angiogenic agents, corticosteroids, NSAIDs, and/or taxane-based chemotherapy, or who have prior history of peptic ulceration or diverticular disease, are at increased risk. OSI-779 should be permanently discontinued in patients who develop gastrointestinal perforation.

Bullous and exfoliative skin disorders: Bullous, blistering, and exfoliative skin conditions have been reported, including very rare cases suggestive of Stevens-Johnson syndrome/toxic epidermal necrolysis (which in some cases were fatal). OSI-779 treatment should be interrupted or discontinued if the patient develops severe bullous, blistering, or exfoliating conditions.

Ocular disorders: Very rare cases of corneal perforation or ulceration have been reported during use of OSI-779. Other ocular disorders, including abnormal eyelash growth, keratoconjunctivitis sicca or keratitis have been observed with OSI-779 treatment and are known risk factors for corneal perforation/ulceration.

OSI-779 therapy should be interrupted or discontinued if patients present with acute/worsening ocular disorders such as eye pain.

Fertility and teratology: These studies have not been conducted with OSI-774, and safety for women of childbearing capacity cannot be implied from the existing data. OSI-774 has not been tested for carcinogenic activity in a lifetime rodent bioassay.

16.1.1.4 Lapatinib

Lapatinib is a small molecule member of the quinazoline class of kinase inhibitors and is a dual reversible inhibitor of both EGFR and HER2 receptors. Lapatinib has a median inhibitory concentration for the EGFR receptor at a dose of 12 nmol/l and HER2 receptor at a dose of 9 nmol/l. Lapatinib prevents phosphorylation and subsequent signal transduction of both the Ras/Raf mitogen activated protein kinase and the phosphoinositol-3-kinase/Akt pathways, leading to an increase in apoptosis and decreased cellular proliferation. Lapatinib at nanomolar concentration blocks cell-cycle proliferation at the G1 phase. It has a high specificity for both the EGFR TKD and HER2 TKD and has no other inhibition of other cell receptors and nonreceptor tyrosine kinases. Results from pharmacokinetic studies showed that lapatinib is highly protein bound in humans (> 99%). Oxidation is the primary route of metabolism via the hepatic cytochromes CYP3A4, CYP3A5 and CYP2C19.

Lapatinib has been approved by the FDA to be used in combination with capecitabine for treatment of patients with metastatic breast cancer whose tumors overexpress HER2. A Phase III trial in women whose tumors overexpressed HER2 and who had failed trastuzumab randomized patients to receive either lapatinib at 1250 mg once daily (continuously) plus capecitabine 2000 mg/m2/day on days 1 – 14 every 21 days, or to receive capecitabine alone at a dose of 2500 mg/m2/day on days 1 – 14 every 21 days. The primary end point of this study was time to progression (TTP). Based on the results of a prespecified interim analysis, further enrollment was discontinued after 399 patients demonstrated an increase in TTP from 19.7 weeks to 36.9 weeks. The combination increased toxicity with notable side effects of diarrhea (65%), hand-foot

syndrome (53%), nausea (44%), rash (29%) and fatigue (24%). QT prolongation is another potential toxicity of lapatinib.

Lapatinib has been evaluated as a single agent in several Phase II studies. It has been studied in inflammatory breast cancer overexpressing HER2. This trial demonstrated a 35% objective response rate (ORR). It has been further investigated as a monotherapy in a Phase II trial involving heavily pretreated metastatic breast cancer patients. In this study no objective responses were seen in patients with HER2 negative tumors. A 4.3% response rate in HER2-positive patients was demonstrated. A third Phase II study involved single-agent lapatinib in HER2-positive breast cancer patients with brain metastases. In the 39 subjects enrolled in this trial, only one had a CNS response.

HER2 overexpression has been reported in 4 – 20% of NSCLC and is associated with a poor prognosis. A Phase II trial tested lapatinib as monotherapy in the first- or second-line setting in metastatic NSCLC. This trial was discontinued after enrollment of 131 patients, after only one PR was seen. The use of lapatinib is now being tested in other solid tumors, including gastric, ovarian and colorectal carcinomas.

Predictors of HER2-TKI sensitivity and resistance

Clinical data have so far demonstrated that tumors must overexpress HER2 to be sensitive to either lapatinib or trastuzumab. Lapatinib sensitivity increases with increased expression of both HER2 and EGFR. In hormone-positive breast cancer, increased HER2 expression has been shown to downregulate estrogen receptor (ER) expression. Signaling through HER2 seems to bypass the estrogen requirement for breast cancer cell growth and may drive initially ER-positive cells into an ER-negative, or endocrine therapy-resistant state. Overexpression of both EGFR and HER2 has been associated with tamoxifen resistance. Estrogen and tamoxifen have been shown to increase phosphorylation of HER2. ER and HER2 have a bidirectional crosstalk; therefore lapatinib may have the ability to restore tamoxifen sensitivity. A Phase I study combining letrozole and lapatinib determined the optimal Phase II dosage of letrozole at 2.5 mg/day and lapatinib at 1500 mg/day, which is being tested at present.

Trastuzumab resistance has been seen in tumors with upregulated insulin-like growth factor receptor-1 (IGF-1R), in tumors that lack phosphatase and tensin homolog (PTEN) expression, and in tumors with activating mutations in PI3K. An overexpression of IGF-1R leads to increased phosphorylation of HER2, resulting in increased signaling. Both PTEN-deficient tumors as well as tumors with activating PI3K mutations have an increase in the PI3K signaling pathway. Additional resistant mechanisms to trastuzumab include increased membrane-associated glycoprotein mucin-4 (MUC4), which interacts directly with HER2. MUC4 provides steric hindrance to trastuzumab binding.

There are limited data defining the exact cause of lapatinib resistance. HER2 receptor tyrosine kinase mutations have rarely been reported in breast cancer. Proposed mechanisms of resistance have included an upregulation of estrogen receptor pathways and overexpression of both HER3 and HER4. Other proposed mechanisms of resistance have included upregulation of IGF-1R and lack of inhibition of surviving.

18.1.2 Other agents

18.1.2.1 Sunitinib Malate

Sunitinib malate (sunitinib; SU11248; SU011248; Sutent®) is an oral, multi-targeted, small molecule inhibitor of the receptor tyrosine kinases (RTKs) involved in tumor proliferation and angiogenesis, including vascular endothelial growth factor receptor-1 (VEGFR-1), -2, and -3, platelet-derived growth factor receptor (PDGFR) -□ and -□, stem cell factor receptor (KIT), the tyrosine kinase (TK) receptor encoded by the ret proto-oncogene (RET; rearranged during transfection), fms-like tyrosine kinase 3 (Flt3), basic fibroblast growth factor (bFGF) and colony-stimulating factor (CSF)-1R (O'Farrell et al., 2003a; Chow and Eckhardt, 2007; Faivre et al., 2007; Gan et al., 2009; Mashkani et al., 2010). Sunitinib selectively and potently inhibits the class III and class V split-domain RTKs (Mendel et al., 2003).

Sunitinib shows significant antitumor and anti-angiogenic activity in a number of human tumor xenograft and angiogenesis models in mice as well as in phase 1 and 2 studies in patients with a variety of tumor types (Gan et al., 2009; Mena et al., 2010). As of June 2008, a total of 8932 subjects with solid malignant tumors have received sunitinib, including patients with renal cell carcinoma (RCC) and those with gastrointestinal stromal tumors (GIST) (Investigator's Brochure, 2009). In phase 2 studies in cytokine-refractory metastatic RCC, sunitinib produced objective responses in 40% of patients with a median time-to-tumor-progression (TTP) of 8.7 months (Motzer et al., 2006). In phase 3 studies of patients with imatinib-resistant GIST, sunitinib was highly superior to placebo ($p<0.0001$) with respect to median TTP (27.3 weeks vs. 6.4 weeks), progression-free survival (PFS), and overall survival (OS) (Demetri et al., 2006).

Sunitinib was granted regular approval on January 26, 2006 by Food and Drug Administration (FDA) for the treatment of gastrointestinal stromal tumor (GIST) after disease progression on or intolerant to imatinib mesylate and accelerated approval for advanced renal cell carcinoma (RCC) (Goodman et al., 2007; Izzedine et al., 2007; Rock et al., 2007), which was changed to regular approval on February 2, 2007.

Mechanism of Action

Tumor VEGF expression has been associated clinically with disease prognosis in many different types of malignancies. VEGF expression is increased by diverse stimuli including proto-oncogene activation and hypoxia, with the hypoxic state frequently arising in solid tumors because of inadequate perfusion. In addition to its angiogenic role, VEGF also profoundly increases the permeability of the vasculature thereby, potentially contributing to tumor progression. A leaky tumor endothelium enhances nutrient and catabolite exchange and represents less of a barrier to tumor cell migration and intravasation during metastasis. Two high-affinity receptors for VEGF with associated TK activity have been identified on human vascular endothelium; VEGFR-1/Flt-1 and VEGFR-2/kinase insert domain-containing receptor (KDR). Although the relative contributions of KDR and Flt-1 signaling in mediating tumor progression have not been elucidated, a number of studies suggest that KDR performs a predominant role.

In addition to VEGF receptor signaling, increasing evidence implicates PDGFR signaling in tumor angiogenesis. Recent nonclinical evidence suggests that inhibition of PDGFR signaling

augments the antitumor and anti-angiogenic effects of VEGFR inhibitors. In addition, PDGF signaling is implicated in the autocrine growth of tumor cells and in the recruitment and regulation of tumor fibroblasts.

Upon chronic oral dosing, sunitinib is expected to inhibit PDGF- and VEGF-driven angiogenesis and as a consequence, limit solid tumor growth. Because angiogenesis is necessary for the growth and metastasis of solid tumors, and VEGF is believed to have a pivotal role in this process, sunitinib treatment may have broad-spectrum clinical utility (Chow and Eckhardt, 2007; Gan et al., 2009). Sunitinib also exerts direct antitumor activity on cells that express target RTKs associated with tumor cell proliferation, such as KIT, PDGFR, and RET. The clinical activity of sunitinib in patients with advanced GIST is an example of this antitumor effect.

Nonclinical Specificity and Efficacy Studies

In vitro studies have demonstrated the specificity of sunitinib for inhibition of the Class 3 and Class 5 RTKs, including receptors for VEGF (VEGFR), KIT, Flt-3, and PDGFR (Investigator's Brochure, 2009). Specifically, receptor phosphorylation inhibition studies have shown that sunitinib inhibits KIT-ligand-induced phosphotyrosine levels in a dose-dependent manner with IC50 values of 0.001-0.01 mcM in vitro and reduced PDGFR-β phosphotyrosine levels in vivo (Abrams et al., 2003a). Sunitinib also selectively inhibited proliferation of human umbilical vein endothelial cells (HUVEC) stimulated with VEGF (IC50=0.04 mcM) compared to FGF-stimulated proliferation (IC50=0.7 mcM) (Mendel et al., 2003).

In animal efficacy studies, sunitinib showed broad antitumor activity in mouse xenograft models against a variety of human tumor cell lines including colorectal cancer (HT-29, Colo205), non-small cell lung cancer (H460), breast cancer (MDA-MB-435), melanoma (A375), epidermoid cancer (A431), and glioma (SF763T) (Mendel et al., 2003). Sunitinib has also demonstrated antitumor activity against other breast cancer models (MMTV-v-Ha-ras transgenic mouse mammary carcinoma and dimethylbenzanthracene [DMBA]-induced rat mammary carcinomas) (Abrams et al., 2003b). In an animal model of KIT-expressing small cell lung cancer (SCLC; NCI-H526), sunitinib administration resulted in greater tumor growth inhibition than did imatinib (Abrams et al., 2003a).

Combination studies of sunitinib with docetaxel, 5-fluorouracil (5-FU), or doxorubicin resulted in significantly enhanced growth inhibition of human MX-1 breast cancer xenografts compared to levels of inhibition with either sunitinib or the cytotoxic agent alone (Abrams et al., 2003b). Moreover, the combination therapies each led to a significantly increased survival compared to either single agent alone (Abrams et al., 2003b). Significantly delayed tumor growth has also been demonstrated in combination studies of sunitinib and cisplatin in NCI-H526 SCLC xenografts (Abrams et al., 2003a).

Additional nonclinical *in vitro* and *in vivo* studies are summarized in Chow and Eckhardt, 2007.

Nonclinical Toxicology Studies

Single- and multiple-dose toxicology studies were conducted in mice, rats, rabbits, dogs, and monkeys (Investigator's Brochure, 2009). The acute oral maximally-tolerated dose (MTD) for mice, rats, and dogs was greater than the maximum dose of 500 mg/kg. The MTD in monkeys was greater than the 1200 mg/kg maximum dose tested, but emesis occurred at doses ≥50 mg/kg.

Treatment-related effects in the lymphoid tissue, bone marrow, adrenal glands, and bone growth plate were seen in rat repeated-dose studies with gastrointestinal tract, reproductive organ, kidney, pancreas, and pituitary effects reported at the highest dose. Death was observed at the highest dose level of 240 mg/kg/day. Gastrointestinal disturbances (diarrhea, loss of appetite, emesis) as well as hematologic disturbances also occurred in an 8-week study in female monkeys. Other toxicities seen in monkeys included mild elevations in AST, ALT, and creatinine kinase (CK), adrenal gland cortex hemorrhage, acinar degranulation of the salivary glands, decreased erythropoiesis in the bone marrow, and lymphoid atrophy. Possible impairment of immune function in the highest dose group was manifested as cytomegalovirus and bacterial infections. There is an indication that repeated high doses of sunitinib may lead to cardiac function/contractility changes as confirmed by altered ECG and MUGA or echocardiographic parameters and increased cTnI and/or T in single animals that died or were euthanized early due to a moribund condition. These changes appear to be primarily functional and reversible. Due to the poor clinical condition of these animals, it appears that the cardiac changes were not a direct result of sunitinib treatment, but rather resulted from an important, noncompensated volume loss and additional suppression of heart function due to chronotropic incompetence and possible myocardial involvement of uncertain etiology. Data from this study indicate that functional cardiac changes were induced primarily at the highest dose of sunitinib.

Sunitinib was found to be negative for genotoxicity in vivo and in vitro. Wound-healing studies showed a subtle transient delay in skin wound healing in mice treated continuously for up to 5 weeks with supratherapeutic doses of sunitinib at 80 mg/kg/day. However, it was determined that this alteration in wound healing has minimal biologic significance.

Nonclinical Pharmacokinetics

Single-dose pharmacokinetics (PK) was evaluated in mice, rats, rabbits, dogs, and monkeys at oral doses of 20, 40, 80, and 160 mg/kg, respectively (Investigator's Brochure, 2009). At these doses, the respective Tmax values in mice were 3, 3, 0.5, and 6 hours, while Cmax values were 420, 708, 877, and 2670 mg/mL, respectively. In monkeys, t1/2□, t1/2□, and t1/2□ were 4, 14.9, and 252 hours, respectively. Repeat- dose PK studies indicated that increases in exposure were not consistently proportional to dose. Steady state plasma concentrations were reached after 28 days of dosing with little change in levels thereafter. Sunitinib appeared to readily distribute into the CNS in mice and to a lesser extent in rats and monkeys.

Sunitinib metabolism is predominantly mediated by CYP3A4 and produces an active metabolite, SU012662. This metabolite and sunitinib were the only major drug-related compounds found in the systemic circulation in mice, rats, monkeys and humans. Sunitinib and its major metabolite are highly (90-98%) protein bound in mouse, rat, monkey, and human plasma. IC50 values measured in vitro are expected to be reached with the currently recommended 50 mg dose. Sunitinib and SU012662 are not potent inducers or inhibitors of major CYP450 enzymes. Therefore, they are both predicted to have a low potential to cause clinically relevant drug-drug interactions mediated by CYP450 enzymes and efflux transporters. However, concurrent treatment with CYP3A4 inducers and inhibitors may affect sunitinib metabolism.

Clinical Experience

As of June 2008, 9204 subjects had received at least one dose of sunitinib in 77 completed or ongoing clinical studies, with 8986 subjects having received multiple doses of the agent (Investigator's Brochure, 2009). In phase 1 studies, sunitinib demonstrated single-agent activity in patients with RCC, GIST, non-GIST sarcomas, non-small cell lung cancer (NSCLC), colorectal cancer, neuroendocrine tumors (NET), melanoma, prostate cancer, and thyroid cancer. Sunitinib has also been studied in the phase 1 setting in patients with acute myeloid leukemia (AML). Pivotal trials of sunitinib in imatinib-resistant GIST (a placebo-controlled phase 3 trial), and metastatic RCC (MRCC) (single-arm, non-randomized, multicenter, open-label trial) and supporting trials in each disease were completed and submitted in support of the New Drug Application (NDA). In addition, there are several ongoing company-sponsored single agent and combination clinical trials for a variety of other indications.

Phase 1 Experience

In an early phase 1 study designed to investigate dosing regimen and scheduling (in human subjects, the results of clinical pharmacology studies demonstrate that Cmax and AUC increased in a proportional manner after single doses of 50 to 350 mg as well as after multiple doses of 25-100 mg), 41 patients with a variety of advanced solid tumors received sunitinib administered on a schedule of 2 weeks of treatment followed by 2 weeks off (2/2 schedule) or 4 weeks on with 2 weeks off (4/2 schedule) (Rosen et al., 2003). Doses evaluated on the 2/2 schedule (n=23) included 50 mg every other day (n=3), 50 mg daily (n=15), or 75 mg daily (n=5); the 18 patients enrolled in the 4/2 schedule received 25 mg daily (n=3) or 50 mg daily (n=15). The most frequent adverse events (AEs) were constitutional (fatigue/asthenia), gastrointestinal (nausea, vomiting, diarrhea) and hematologic (neutropenia, thrombocytopenia). Most of the AEs were grade 1 or 2, although at 75 mg daily, grade 3 and 4 fatigue/asthenia were dose limiting but readily reversible on discontinuation of treatment. There were 4 partial responses (PRs) assessed by RECIST and 22 patients with stable disease (SD) among the 41 patients.

A phase 1 dose-escalation study in 28 patients with advanced tumors evaluated sunitinib doses of 30 mg/m2 every other day, and doses of 30, 42, or 59 mg/m2 daily on the 4/2 schedule (Faivre et al., 2006). Grade 3 fatigue and hypertension were dose limiting at 59 mg/m2 as well as grade 2 bullous skin toxicity, and the MTD was defined as 42 mg/m2 daily. Based on these and other reversible AEs in the 12 patients treated at the MTD, the recommended phase 2 dose on the 4/2 schedule was determined to be 50 mg/day. Responses determined by RECIST were seen in 6 of 23 evaluable patients: 3 in RCC, 1 in NET, 1 in GIST, and 1 in adenocarcinoma of unknown primary. Tumor responses in patients treated at higher doses were often associated with reduced intratumoral vascularization and central tumor necrosis, leading to organ perforation in one patient and fistula in another. These observations suggest the possible necessity for careful tumor density monitoring to detect early evidence of necrosis.

Two phase 1 studies have been conducted in AML, the first with the primary endpoint of evaluation of the inhibition of FLT3 phosphorylation (O'Farrell et al., 2003b) and the second designed as a conventional dose-escalation study (Fiedler et al., 2005). O'Farrell and colleagues studied FLT3 phosphorylation in 29 AML patients who received a single dose of sunitinib at doses ranging from 50-350 mg. Over 50% of patients showed strong inhibition of Flt3 phosphorylation at doses of 200 mg and higher. As anticipated from nonclinical data, patients

with FLT3 internal tandem duplication (ITD) mutations were more sensitive than those with wild-type Flt3 (FLT3-WT) as shown by 100% inhibition in FLT3-ITD compared to 50% in FLT3-WT. This study also gave evidence of downstream signal inhibition (STAT5 and ERK pathways), with STAT5 levels reduced primarily in FLT3-ITD patients while ERK inhibition occurred in the majority of patients independently of FLT3 inhibition. The dose-escalation study enrolled 15 patients with refractory or resistant AML who were treated with sunitinib on either the 4/2 or 4/1 schedule at a starting dose of 50 mg/day (Fiedler et al., 2005). Dose-limiting AEs (grade 4 fatigue and hypertension) occurred in both patients treated at 75 mg/day, and one of these patients (who had received prior mitoxantrone) developed cardiac failure. The 75 mg dose level was therefore terminated and 50 mg/day was considered to be the MTD. All four patients with FLT3 mutations had morphologic or partial responses compared to 2 of 10 evaluable patients with wild-type FLT3. Responses, although longer in patients with mutated FLT3, were of short duration.

Preliminary results from phase 1 studies exploring the combination of sunitinib with chemotherapeutic agents like capecitabine (Sweeney et al., 2007; Chiorean et al., 2008; Royce et al., 2008), pemetrexed (Chow et al., 2008), docetaxel (Traynor et al., 2008), gemcitabine (Brell et al., 2008; Michaelson et al., 2008), FOLFOX (Leong et al., 2007), FOLFIRI (Starling et al., 2008), carboplatin/paclitaxel (Heath et al., 2008), and metronomic cyclophosphamide/methotrexate (Rugo et al., 2008) in patients with various solid tumors have been presented. The MTD of sunitinib on the 4/2 schedule with docetaxel (60 mg/m2) was 25 mg daily; with capecitabine (1000 mg/m2), it was 37.5 mg daily. The MTD of sunitinib on the 2/1 schedule with docetaxel (75 mg/m2) was 37.5 mg daily and with capecitabine (1000 mg/m2) was 50 mg daily. The MTD for sunitinib as continuous daily dosing (CDD) with capecitabine (1000 mg/m2) was 37.5 mg daily and for pemetrexed (500 mg/m2) was 37.5 mg daily. Reported DLTs included febrile neutropenia, fatigue, hand-foot syndrome, gastrointestinal hemorrhage, cerebral hemorrhage, and ischemic optic neuropathy. Phase 1 trials evaluating the combination of sunitinib with other targeted agents like temsirolimus (Patel et al., 2009; Fischer et al., 2008), bevacizumab (Feldman et al., 2008; Cooney et al., 2008) and IFN- α (Motzer et al., 2009a) have shown problems with increased toxicity in patients with RCC (Feldman et al., 2008, Fischer et al., 2008; Motzer et al., 2009a). However, the combination of bevacizumab with sunitinib in patients with miscellaneous solid tumors was well tolerated (Cooney et al., 2008).

Phase 2 and 3 Experience

Updated results have recently been published on 750 patients with MRCC treated on a phase 3 study of sunitinib at a daily dose of 50 mg on the 4/2 schedule compared to interferon (IFN)-α 9 MU subcutaneously thrice weekly (Motzer et al., 2009b). Median overall survival (OS) was greater in the sunitinib group than in the IFN-α group (26.4 vs. 21.8 months, respectively). Sunitinib treatment was associated with a higher objective response rate (RR) than IFN-α (47% vs. 12%, respectively). Eleven patients in the sunitinib group and four patients in the IFN-α group achieved a complete response per investigator assessment. Median progression-free survival (PFS) was 11 months for sunitinib compared with 5 months for IFN-α. The most commonly reported sunitinib-related grade 3 adverse events included hypertension (12%), fatigue (11%), diarrhea (9%), and hand-foot syndrome (9%). An exploratory analysis, which censored 25 patients from the IFN-α group who had crossed over to receive sunitinib on study,

showed a median OS time of 26.4 months for sunitinib compared with 20 months for the IFN-α group. Results from a phase 1/2 dose-finding trial of sunitinib plus gefitinib, enrolling 42 patients, have been published (Motzer et al., 2010). In phase 1, patients received sunitinib 37.5 or 50 mg on the 4/2 schedule plus gefitinib 250 mg, both once daily. The MTD was determined to be 37.5 mg. Two DLTs were observed with the 50 mg dose: grade 2 left ventricular ejection fraction decline and grade 3 fatigue. In phase 2, patients received sunitinib at the MTD plus gefitinib. Thirteen patients treated at the MTD achieved a partial response and 12 had stable disease. Median PFS was 11 months. The most commonly reported grade 3/4 adverse event was diarrhea.

The promising results in phase 1 trials and the involvement of KIT and PDGFR-α, two of the sunitinib target RTKs in GIST led investigators to undertake a phase 1/2 trial in patients with GIST refractory or intolerant to imatinib to determine an appropriate dose and regimen for phase 2 development of sunitinib in this disease. Patients received up to 75 mg sunitinib daily on the 2/2, 4/2, or 2/1 schedule, with 50 mg daily on the 4/2 schedule being selected for continued study. In all, 75 patients were treated on the trial. Among 41 patients treated for at least 6 months, 6 had an objective response (OR; RECIST criteria) and an additional 16 had cessation of disease progression and minor responses for >6 months. Overall, 54% of the 41 patients had evidence of clinical benefit (OR or PFS) (Desai et al., 2004). Determination of the GIST genotype in these 41 patients showed that clinical benefit had been achieved in several secondary mutational variants that conferred imatinib resistance (Demetri et al., 2004). Fifty-three of the GIST patients treated on this trial subsequently underwent serial 18FDG-PET imaging where qualitative responses were graded as good in 33/53 patients, mixed in 15/53, and poor in 5/53 (Dileo et al., 2005). Correlation with clinical response (Fisher's exact p=0.03) showed that 22 of 33 patients graded as good by 18FDG-PET imaging had clinical benefit (OR or SD ≥ 6 months) after 6 months of therapy while 4 of 15 patients graded as mixed had benefit as did 2 of 5 patients graded as poor.

Data from a pivotal multinational, randomized (2:1), double-blind, placebo-controlled phase 3 trial in over 300 patients with imatinib-resistant GIST has shown significant clinical effect with sunitinib compared with placebo (Demetri et al., 2006). Patients on the active treatment arm received sunitinib at a dose of 50 mg daily on the 4/2 schedule. The median TTP for the treatment arm (n=207) was 27.3 weeks compared to a median of 6.4 weeks for placebo (n=105) (p<0.0001). Therapy was reasonably well tolerated; the most common adverse events were fatigue, diarrhea, skin discoloration and nausea.

In addition to patients with RCC and GIST, sunitinib has been evaluated in breast cancer (Burstein et al., 2008), NSCLC (Socinski et al., 2008), transitional cell carcinoma (TCC) (Gallagher et al., 2008), NET (Kulke et al., 2008), thyroid carcinoma (Cohen et al., 2008), certain subtypes of sarcoma (Keohan et al., 2008; Vigil et al., 2008), gastro-eosophageal cancer (Moehler et al., 2009), high-grade glioma (Chaskis et al., 2008), squamous cell carcinoma of the head and neck (SCCHN) (Choong et al., 2008), hepatocellular carcinoma (HCC) (Zhu et al., 2008), colorectal carcinoma (CRC) (Saltz et al. 2007; Starling et al., 2008; Pfeiffer et al., 2009) and uveal melanoma (Chan et al., 2008). Results from these trials have been summarized in Gan et al., 2009.

Safety Profile

Sunitinib is reasonably well tolerated, with asthenia, hypertension, dermatitis, and mild myelosuppression as the most common AEs (Stadler, 2006). Additionally, the inhibition of TK receptors by agents such as sunitinib can result in cutaneous AEs such as acral erythema, subungal splinter hemorrhages, modification of hair and skin pigmentation, mucositis, and (occasionally) periocular edema (Robert et al., 2005; Suwattee, 2008). Hand-foot skin reaction, a group of signs and symptoms that can affect, usually bilaterally, the hands and/or feet of patients, has occurred in patients receiving sunitinib (Porta et al., 2007; Suwattee, 2008). A recent analysis of dermatological AEs in patients receiving sunitinib therapy has reported that all-grade hand-foot skin reactions occurred in 19% of patients (5% grades 3-4), skin discoloration in 28% (no grades 3-4), dry skin in 16% (1% grades 3-4), skin rash in 13% (1% grades 3-4), dermatitis in 8% (2% grades 3-4), hair color changes in 10% (no grades 3-4), alopecia in 6% (no grades 3-4), and phototoxicity in <0.1% (no grades 3-4) (Rosenbaum et al., 2008).

Cardiotoxicity, including congestive heart failure (3%-8%) and left ventricular dysfunction (12%-14%), has been reported in patients undergoing treatment with sunitinib (Chu et al., 2007; Khakoo et al., 2008; Schmidinger et al., 2008; Telli et al., 2008). More subjects treated with sunitinib experienced decline in left ventricular ejection fraction (LVEF) than subjects receiving either placebo or IFN-α (Investigator's Brochure, 2009). In a phase 3 GIST study in subjects with imatinib-resistant or -intolerant GIST, 22 of 209 (11%) subjects on sunitinib and 3 of 102 (3%) subjects on placebo had treatment-emergent LVEF values below the lower limit of normal (LLN). In a phase 3 study treatment-naive RCC patients, 27% and 15% of subjects on sunitinib and IFN-α, respectively, had an LVEF value below the LLN. Among 461 patients enrolled in CDUS-monitored trials with sunitinib alone, 2% of patients experienced left ventricular systolic dysfunction (CDUS data). It is unknown whether patients with concurrent cardiac conditions may be at a higher risk for developing drug-related LVEF. Baseline and periodic evaluations of LVEF should be considered while these patients are on sunitinib treatment. In patients without cardiac risk factors, a baseline evaluation of LVEF should be considered.

Sunitinib has been shown to prolong the QT interval in a dose-dependent manner, which may lead to an increased risk for ventricular arrhythmias, including Torsade de Pointes. This condition has been reported in <0.1% of sunitinib-exposed patients. The DCTD, NCI, issued an IND AE Action Letter to all investigators using sunitinib describing the occurrence of QTc prolongation and Torsade de pointes (ventricular tachycardia) in patients on clinical trials utilizing sunitinib. As a result of this Action Letter, DCTD, NCI-sponsored sunitinib protocols were amended to include the requirement for a baseline EKG prior to study treatment, exclude patients with histories of serious ventricular arrhythmias or prolonged QTc, and exclude patients with certain cardiac conditions.

Among 8054 solid tumor subjects treated with single-agent sunitinib, Grade 3 hypertension was reported in 6.1% of patients and was one of the most commonly reported Grade 3 AEs. Of subjects receiving sunitinib for treatment-naive MRCC, 34% receiving sunitinib experienced hypertension, compared with 4% on IFN-α. Grade 3 hypertension was reported in 13% of treatment-naive metastatic RCC subjects on sunitinib compared to <1% on IFN-α. Severe

hypertension occurred in 4% GIST subjects on sunitinib, 1% GIST subjects on placebo, 9% RCC subjects on sunitinib and 1% RCC subjects on IFN-α (Investigator's Brochure, 2009). Among 461 patients enrolled in CDUS-monitored trials with sunitinib alone, 28% experienced hypertension (CDUS data).

In subjects receiving sunitinib for treatment-naive MRCC, 37% had bleeding events compared with 8% receiving IFN-α (Investigator's Brochure, 2009). Bleeding events occurred in 18% of patients receiving sunitinib in the double-blind treatment phase of the GIST phase 3 study, compared with 17% receiving placebo. Among 461 patients enrolled in CDUS-monitored trials with sunitinib alone or in combination with other agents, there were 103 reported bleeding events (CDUS data). Epistaxis was the most common hemorrhagic AE reported. Tumor-related hemorrhage can occur with sunitinib and in the case of pulmonary tumors, may present as severe and life-threatening hemoptysis or pulmonary hemorrhage.

Hypothyroidism has been reported in 71% patients with RCC (Rini et al., 2007) and in 36% of those with GIST (Desai et al, 2006) treated with sunitinib. Among subjects treated with single-agent sunitinib, hyperthyroidism was reported in 0.6% and thyroiditis in 0.1% (Investigator's Brochure, 2009). Baseline measurement of thyroid function is recommended, and patients with thyroid dysfunction should be treated appropriately prior to starting sunitinib therapy.

Nonclinical evidence of adrenal toxicity following sunitinib exposure led the company to perform specialized safety assessments in clinical studies, including computed tomography or MRI in 336 subjects to specifically identify any change in adrenal gland structure or the presence of adrenal gland hemorrhage (Investigator's Brochure, 2009). Neither event was observed. Adrenocorticotropic hormone (ACTH) stimulation testing was done in 400 patients across multiple sunitinib trials. One subject developed consistently abnormal test results during treatment that were unexplained and may be related to sunitinib treatment. Eleven additional subjects had abnormailities in the final test, with low peak cortisol levels. None of these patients had clinical evidence of adrenal insufficiency. However, based on the nonclinical findings, patients receiving sunitinib should be clinically followed for signs and symptoms of adrenal insufficiency, especially in (1) patients with comorbidities associated with adrenal dysfunction, (2) patients with preexisting adrenal insufficiency (primary or secondary), and (3) patients with concomitant stress (e.g., fever, infection, bleeding, serious accident, surgery) that may precipitate overt adrenal insufficiency in the presence of subclinical sunitinib-induced adrenal toxicity.

Diarrhea, nausea, abdominal pain, vomiting, constipation and dyspepsia are some of the most frequent AEs reported with sunitinib (Investigator's Brochure, 2009). Serum chemistries including phosphate should be performed at the beginning of each treatment cycle. Supportive care may include anti-emetic premedication, supportive oral care products, and analgesics. Serious complications due to degeneration or shrinkage of tumors, including gastrointestinal perforation and tracheoesophageal fistula, have occurred rarely in patients with abdominal, head and neck, thyroid, and other malignancies treated with sunitinib, believed to be the result of the antitumor effect of sunitinib. Other serious effects include thromboembolic events, rare reversible posterior leukoencephalopathy syndrome (RPLS), proteinuria with rare nephrotic syndrome, and rare microangiopathic hemolytic anemia.

Clinical Pharmacokinetics

Orally-administered sunitinib is well absorbed in humans, with linear pharmacokinetics (PK) at doses of 50-150 mg/day (Sakamoto, 2004). Sunitinib is highly protein-bound and metabolized through N-de-ethylation by cytochrome P450 (CYP) 3A4 to SU12662 (Investigator's Brochure, 2009). SU12662 has a similar inhibitory profile to sunitinib in vitro and similar protein binding properties. SU12662 is further metabolized by CYP3A4 to a minor inactive metabolite (Investigator's Brochure, 2009).

The PK of sunitinib was studied in a variety of company-sponsored studies in both healthy volunteers (n=135) and in patients with solid tumors (n=266), including GIST and metastatic RCC; the PK was similar in the volunteers and in those with solid tumors. Terminal half-lives (t1/2) of sunitinib and SU12662 are 40-60 hours and 80-110 hours, respectively, with a time to maximum concentration (Tmax) of 6-12 hours for sunitinib and its primary active metabolite, followed by a biexponential decline in concentrations. The PK of sunitinib and SU12662 were measured in a phase 1 dose-escalation study in patients with advanced solid malignancies (Faivre et al., 2006). Twenty-eight patients received doses ranging from 15 mg/m2 to 59 mg/m2 (ranging from 50 mg every other day to 150 mg/day), on a 4 weeks on, 2 weeks off (4/2) schedule. Concentration-versus-time data were analyzed using a noncompartmental analytic technique. Overall, sunitinib displayed a long half-life and a large volume of distribution with moderate interpatient variability. Trough plasma concentrations of sunitinib and SU12662 increased with increasing doses. However, area under the concentration-time curve (AUC) values increased less than proportionally with dose. Accumulation ratios of sunitinib were >1, with detectable trough drug levels, suggesting drug accumulation over time. At the recommended dose, maximum plasma concentration (Cmax) occurred approximately 5 hours after administration and t1/2 ranged from 41-86 hours. Doses of 50 mg daily led to plasma concentrations ranging from 50-100 ng/mL. Most patients with DLTs had combined (sunitinib plus SU12662) trough plasma concentrations ≥100 ng/mL (Faivre et al., 2006).

PK values determined from body surface area (BSA)-based doses were adjusted to reflect fixed doses of 50, 75-100, and 100-150 mg doses to determine if there was a need for BSA-based dosing to be employed. AUC sum values obtained using BSA-normalized and fixed dosing were found to be comparable, suggesting that normalizing the dose based on BSA would not improve variability. Therefore, fixed dosing on a milligram basis was considered appropriate for phase 2 studies (Faivre et al., 2006).

To determine the effect of food on the PK of sunitinib and its active metabolite SU12662, 16 healthy subjects received a single dose of sunitinib 50 mg under fasting conditions and 14 subjects received a single dose of sunitinib 50 mg under fed conditions (Bello et al., 2006). Subjects were randomized to one of two treatment sequences each comprising two treatment periods (fasted and fed). In Sequence 1, the fasted treatment period was followed by the fed treatment period, and for Sequence 2, the fasted period followed the fed period. For the fasted period, a single oral (PO) dose of sunitinib 50 mg was administered after a 10-hour fast. For the fed period, a single PO dose of sunitinib 50 mg was administered within 30 minutes of a high-fat, high-calorie meal. A washout period of at least 4 weeks separated sunitinib dosing between the two treatment periods. Two subjects in Sequence 1 discontinued prematurely due to grade 1 and 2 rash, but PK information was collected and included in the analysis. Only a negligible difference in Tmax of sunitinib was observed between fed and fasted treatment periods;

SU12662 Tmax was prolonged by 2 hours (median difference) in the fed compared with the fasted state. The 90% confidence intervals (CIs) for Cmax and AUC were within the 80-125% bioequivalence range, indicating the absence of food effect. Sunitinib exposure increased slightly in the fed compared with the fasted state (ratios of fed/fasted geometric least square means: Cmax 104%, AUC0-last and AUC0-∞ both 112%). There was a delay in the formation/absorption of the active metabolite SU12662 in the fed state (mean Cmax decreased 23%), but exposure remained unaffected (90% CIs for AUC 0-last and AUC0-∞ were within 80-125%). Sunitinib and SU12662 half-lives, and oral clearance of sunitinib, were not affected by food (Bello et al., 2006).

A study of sunitinib PK in patients with AML indicated that a plasma concentration of 50-100 ng/mL of combined sunitinib and SU12662 could be achieved on the first cycle of a 50 mg/day 4/2 regimen, similar to that achieved in studies with patients having other tumor types (Fiedler et al., 2005).

In a phase 1 trial of sunitinib given on the 4/2 schedule in pediatric patients with relapsed or refractory solid tumors, the median day 21 steady-state trough sunitinib plasma concentration was 24.6 ng/mL (range, 6.0-37.7 ng/mL) at the 15 mg/m2 dose and 37.4 (range, 24.2-62.9 ng/mL) at the 20 mg/m2 dose (DuBois et al., 2008).

Population PK methods indicated that the covariates of weight, gender, race, ethnicity, ECOG score, and tumor type had no clinically significant effects on drug exposure, and that adjustments of starting doses based on these covariates were not required (Investigator's Brochure, 2009).

Concurrent administration of a single dose of sunitinib with ketoconazole (a CYP3A4 inhibitor) in healthy volunteers resulted in 49% and 51% increases in the combined (sunitinib + SU12662) Cmax and AUC0-∞ values, respectively, compared with sunitinib alone (Washington et al., 2003). Concurrent administration of sunitinib and rifampin (a potent CYP3A4 inducer) in healthy subjects resulted in 23% and 46% reduction in combined Cmax and AUC0-∞, respectively, compared with sunitinib alone (Bello et al., 2005). Thus, dose adjustments for sunitinib should be considered when coadministered with CYP3A4 inhibitors and inducers.

Proposed Dose and Schedule for Phase 2 Clinical Trials

Starting doses in multiple-dose studies were 25, 50, 75, and 100 mg administered orally once daily with the majority of patients receiving the 50-mg dose. Patients in sunitinib studies have been treated on four different schedules: schedules 4/1 and 4/2 comprised 4 consecutive weeks of daily dosing followed by a 1- or 2-week rest period, respectively, while schedules 2/1 and 2/2 comprised 2 consecutive weeks of daily dosing followed by a 1- or 2-week rest period, respectively. The majority of subjects were treated on schedules 4/2 or 2/2 in phase 1 studies. Schedule 4/2 has been well tolerated with generally mild to moderate adverse effects at a 50 mg daily dose. Alternate regimens of sunitinib are being explored; 50 mg of sunitinib daily for 2 weeks followed by a 1-week off-treatment period (sunitinib 2/1) (Britten et al., 2008), while another schedule is sunitinib 37.5 mg as a CDD (George et al., 2008). Both schedules were well-tolerated, achieved therapeutic plasma concentrations and showed no significant accumulation of the drug.

18.2 Appendix B: Medication Diaries

CTEP-assigned Protocol #_____

18.2.1 AZD6244 hydrogen sulfate

PATIENT'S MEDICATION DIARY

Today's date _____ Agent_____AZD6244_____

Patient Name_____(initials acceptable) Patient Study ID_____

1. Complete one form for each cycle of treatment.
2. AZD6244 should be stored at room temperature.
3. You will take AZD6244 by mouth twice each day about 12 hours apart. Take the medicine on an empty stomach (no food or drink other than water for 1 hour before or 2 hours after dosing).

   Morning dose: take _____ mg (____capsules) AZD6244. ********** Evening dose: take _____ mg (____capsules) AZD6244.
4. Record the date, the number of AZD6244 capsules you swallowed in the morning and again in the evening, and when you swallowed the medicine.
5. Record the daily dose onto the diary, including missed, skipped, or vomited doses. If you vomit after taking the tablets, the dose is replaced only if the tablets can actually be seen and counted. If you miss a dose, you should resume with the next scheduled dose.
6. If you have any comments or notice any side effects, please record them in the Comments column.
7. Please bring this form and your bottles of AZD6244 when you return for each appointment.

Day	Date	Time of morning dose	Dose taken	Time of evening dose	Dose taken	Comments
			# of capsule taken		# of capsule taken	
1						
2						
3						
4						

5						
6						
7						
8						
9						
10						
11						
12						
13						
14						
15						
16						
17						
18						
19						
20						
21						

Patient's signature _____

Physician's Office will complete this section:

1. Date patient started protocol _____
2. Date patient was removed from _____
3. Patient's planned total daily dose_____
4. Total number of capsules taken this month _____

5. Physician/Nurse/Data Manager's Signature _____

CTEP-assigned Protocol #_____

18.2.2 MK-2206

PATIENT'S MEDICATION DIARY

Today's date _____ Agent: MK-2206

Patient Name_____(initials acceptable) Patient Study ID_____

INSTRUCTIONS TO THE PATIENT:

1. Complete one form for each cycle of treatment.
2. You will take MK-2206 tablets by mouth once every week. You should take the tablets 2 hours before or after a meal.
3. Dose: take _____5 mg tablet(s), _____25 mg tablet(s), and/or _____ 200 mg tablet(s) once every week.
4. Record the date, the number of tablets of each size of tablet that you took, and when you took them.
5. Record the daily dose onto the diary, including missed, skipped, or vomited doses. If you vomit after taking the tablets, the dose is replaced only if the tablets can actually be seen and counted. If you miss a dose, you should resume with the next scheduled dose.
6. If you have any comments or notice any side effects, please record them in the Comments column.
7. Please bring this form and your bottles of MK-2206 tablets when you return for each appointment.

Day	Date	Time of dose	# of tablets taken			Comments
			5 mg	25 mg	200 mg	
1						
2-7						Days 2-7 drug holiday
8						
9-14						Days 9-14 drug holiday
15						
16-21						Days 16-21 drug holiday
22						
23-28						Days 23-28 drug holiday

Patient's signature

_____

Physician's Office will complete this section:

1. Date patient started protocol treatment _____
2. Date patient was removed from study _____
3. Patient's planned total daily dose _____
4. Total number of tablets taken this month _____
5. Physician/Nurse/Data Manager's Signature _____

CTEP-assigned Protocol #_____

18.2.3 Sunitinib

PATIENT'S MEDICATION DIARY

Today's date _____ Agent: Sunitinib

Patient Name_____ (initials acceptable) Patient Study ID _____

INSTRUCTIONS TO THE PATIENT:

1. Complete one form for each 4 week-period while you take sunitinib.
2. You will take your dose of sunitinib by mouth each day in the morning. You will take ____12.5 mg capsule(s), and/or ____ 25 mg capsule(s); or ___ 50 mg capsule. You may take the capsules with or without food as you wish.
3. Record the date, the number of capsules of each size you took, and when you took them.
4. Record the daily dose onto the diary, including missed, skipped, or vomited doses. If you vomit after taking the tablets, the dose is replaced only if the tablets can actually be seen and counted. If you miss a dose, you should resume with the next scheduled dose.
5. If you have any comments or notice any side effects, please record them in the Comments column.
6. Wash your hands with soap and water after touching the capsule(s). Do not share this medication with anyone.
7. Please return unused sunitinib capsules (or empty bottles) and this form to your physician when you go for your next appointment.

Day	Date	Time of daily dose	# of capsules taken			Comments
			12.5 mg	25 mg	50 mg	
1						
2						
3						
4						
5						
6						
7						
8						

9						
10						
11						
12						
13						
14						
15						
16						
17						
18						
19						
20						
21						
22						
23						
24						
25						
26						
27						
28						
29-42						Days 29-42 drug holiday

Patient's signature _____

Physician's Office will complete this section:

1. Date patient started protocol treatment _____
2. Date patient was removed from study _____
3. Patient's planned total daily dose _____
4. Total number of tablets taken this month _____
5. Physician/Nurse/Data Manager's Signature _____

CTEP-assigned Protocol #_____

18.2.4 Erlotinib

PATIENT'S MEDICATION DIARY

Today's date _____ Agent: Erlotinib

Patient Name_____(initials acceptable) Patient Study ID _____

INSTRUCTIONS TO THE PATIENT:

1. Complete one form for each cycle of treatment.
2. You will take theErlotinib tablets by mouth once every day. You will take ____ 25mg tablets, ____100mg tablet, ____150mg tablet. You should take the tablets either 1 hour before or two hours after a meal. Take the tablets with 8 ounces of water.
3. Record the date, the number of tablets of each size of tablet that you took, and when you took them.
4. Record the daily dose onto the diary, including missed, skipped, or vomited doses. If you vomit after taking the tablets, the dose is replaced only if the tablets can actually be seen and counted. If you miss a dose, you should resume with the next scheduled dose.
5. If you have any comments or notice any side effects, please record them in the Comments column.
6. Please bring this form and your bottles of Erlotinib tablets when you return for each appointment.

Day	Date	Time of dose	# of tablets taken			Comments
			mg	mg	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

_____

Physician's Office will complete this section:

1. Date patient started protocol treatment _____
2. Date patient was removed from study _____
3. Patient's planned total daily dose _____
4. Total number of tablets taken this month _____
5. Physician/Nurse/Data Manager's Signature _____

CTEP-assigned Protocol #_____

18.2.5 Lapatinib

PATIENT'S MEDICATION DIARY

Today's date _____ Agent: Lapatinib

Patient Name_____(initials acceptable) Patient Study ID _____

INSTRUCTIONS TO THE PATIENT:

1. Complete one form for each cycle of treatment.
2. You will take Lapatinib tablets by mouth once every day. You should take the tablets 1 hours before or after a meal

   Dose: take ___ mg tablets.
3. Record the date, the number of tablets of each size of tablet that you took, and when you took them.
4. Record the daily dose onto the diary, including missed, skipped, or vomited doses. If you vomit after taking the tablets, the dose is replaced only if the tablets can actually be seen and counted. If you miss a dose, you should resume with the next scheduled dose.
5. If you have any comments or notice any side effects, please record them in the Comments column.
6. Please bring this form and your bottles of Lapatinib tablets when you return for each appointment.

Day	Date	Time of dose	# of tablets taken			Comments
			mg	N/A	N/A	
1						
2						
3						
4						
5						
6						
7						
8						
9						
10						

11						
12						
13						
14						
15						
16						
17						
18						
19						
20						
21						

Patient's signature

_____

Physician's Office will complete this section:

1. Date patient started protocol treatment _____
2. Date patient was removed from study _____
3. Patient's planned total daily dose _____
4. Total number of tablets taken this month _____
5. Physician/Nurse/Data Manager's Signature _____

18.3 Appendix C: 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.

18.4 Appendix D: New York Heart Association Classifications

Clinical Evaluation of Functional Capacity of Patients with Heart Disease in Relation to Ordinary Physical Activity

Class	Cardiac Symptoms	Limitations	Need for Additional Rest*	Physical Ability to work**__
I	None	None	None	Full time
II	Only moderate	Slight	Usually only slight or occasional	Usually full time
III	Defined, with less than ordinary activity	Marked	Usually moderate	Usually part time
IV	May be present even at rest, and any activity increases discomfort	Extreme	Marked	Unable to work

* To control or relieve symptoms, as determined by the patient, rather than as advised by the physician.

** At accustomed occupation or usual tasks.

Reference: Bruce, R. A.: Mod. Concepts Cardiovasc. Dis. 25:321, 1956.

(Modified from New York Heart Association, 1953).

18.5 Appendix E: MEDICATIONS THAT MAY CAUSE QTc PROLONGATION

The following table presents a list of drugs that prolong, may prolong or are unlikely to prolong the QTc. These drugs are prohibited during the treatment with some study agents. Please note that this list is frequently updated. For the most current list of medications, users should be directed to the following website: http://www.azcert.org/medical-pros/drug-lists/drug-lists.cfm.

Compound	Compound Half Life	Possible Washout Period - Hours	Possible Washout Period - Days
Alfuzocin	~10 hours		7
Amantadine	17 +/- 4 hours (10-25)		4
Amiodarone (cordarone)	58 days (15-142) 36 days (active metabolite)		180
Amitriptyline*	> 24 hours, wide interpatient variability		
Arsenic trioxide	Not characterized		
Azithromycin	40 hours		
Bepridil	42 hr (26-64)		10
Chloral hydrate	Readily converted to Trichloroethanol (active metabolite T1/2=7-10 hour)	48	
Chloroquine	Prolonged (days to weeks)		
Chlorpromazine	30 +/- 7 hours		7
Cisapride	6 – 12 hour, up to 20 hour	60	
Clarithromycin	Non linear PK3-4 hr (250mg Q12) 5-7 hr (500mg Q12)	36	
Cloroquine	6 to 60 days; mean 20 days		
Desipramine*	> 24 hours, wide interpatient variability		
Disopyramide	6.7 hr (4-10)	36	
Dofetilide	10 hr	48	
Dolesetron	8.1 hr		
Domperidone	7-8 hr	48	
Doxepin*	> 24 hours, wide interpatient variability		
Droperidol	2.2 hours	10	
Erythromycin	* Each salt form has different Half life*		
Felbamate	20-23 hr		5
Flecainide	20 hr (12-27)		5
Foscarnet	87.5+/-41.8 hours *distribution and release from bone*		20

Fosphenytoin	12-29 hr		6
Gatifloxacin	7-14 hr	48	
Gemifloxacin	7 hours	48	
Grepafloxacin	16 hr		3
Halofantrine	6-10 days (variable among individual)		45
Haloperidol	18 +/-5 hr		5
Ibutilide	6 hours (2-12) * variable among subject*	36	
Imipramine*	> 24 hours, wide interpatient variability		
Indapamide	14 hours (biphasic elimination)		3
Isradipine	8 hours (multiple metabolites)	48	
Levofloxacin	6-8 hours	48	
Levomethadyl	Multiple compartment PK with active metabolite 2.6 day for LAAM, 2 day for nor-LAAM, 4 day for dinor-LAAM		20
Lithium	24 hour (10-50)		7
Compound	Compound Half Life	Possible Washout Period - Hours	Possible Washout Period - Days
Mesoridazine	24-48 hours (animal study)		10
Methadone	15-30 hours		7
Moexipril/HCTZ	2-9 hour (include active metabolite) for moexipril; 5.6-14.8 hours for HCTZ	48	
Moxifloxacin	12 +/-1.3 hours	72	
Naratriptan	6 hours	36	
Nicardipine	~ 2 hour post IV infusion	12	
Nortriptyline*	> 24 hours, wide interpatient variability		
Octreotide	1.7 hours	12	
Ofloxacin	5 to 7.5 hours		2
Ondansetron	4 hours (IV/IM); 3 hours (PO)		1 to 3
Pentamidine	6.4+/-1.3 hours	36	
Pimozide	55 hours		10
Procainamide	3-4 hour for PA and NAPA (active metabolite)	24	
Protiptyline*	> 24 hours, wide interpatient variability		
Quetiapine	6 hours	36	

Quinidine	6-8 hours in adult; 3-4 hours in children	36	
Quinine	4-5 hours		
Risperidone	3-20 hours (extensive to poor metabolizer) 9-hydroxyrisperidone (active metabolite) T ½ =21-30 hours (extensive to poor metabolizer)		4
Salmeterol	5.5 hours (only one datum)	36	
Sotalol	12 hours	72	
Sparfloxacin	20 hours (16-30)		4
Sumatriptan	2.5 hours	12	
Tacrolimus	~34 hours in healthy; ~19 hours in Kidney transplant		7
Tamoxifen	5-7 days (biphasic)		30
Telithromycin	2-3 hr	24	
Thioridazine	20-40 hours (Phenothiazines)		7
Tizanidine	2.5 hours	12	
Vardenifil	4 to 5 hours		
Venlaflaxine	5 +/-2 hours for parent comp. 11+-2 hours for OVD (active metabolite)	60	
Voriconizole	6 hours; dose dependent		
Ziprasidone	7 hr	36	
Zolmitriptan	2.8-3.7 hours (higher in female)	18	

* Weakly associated with Torsades de Pointes and/or QT prolongation but that are unlikely to be a risk for Torsades de Pointes when used in usual recommended dosages and in patients without other risk factors (e.g., concomitant QT prolonging drugs, bradycardia, electrolyte disturbances, congenital long QT syndrome, concomitant drugs that inhibit metabolism).

References:

1. Physician's Desk Reference 2002
2. Facts and Comparisons (update to June 2005)
3. The Pharmacological Basis of Therapeutics 9th Edition, 1996

18.6 Appendix F: LIST OF CYP3A4 INHIBITORS AND INDUCERS

CYP3A4 Inhibitors

HIV Antivirals:	aprepitant	delaviridine
indinavir	erythromycin	diethyl-dithiocarbamate
nelfinavir	fluconazole	fluvoxamine
ritonavir	grapefruit juice	gestodene
	verapamil[2]	imatinib
clarithromycin	diltiazem	mibefradil
itraconazole[1]	cimetidine	mifepristone
ketoconazole[1]	amiodarone	norfloxacin
nefazodone	NOT azithromycin	norfluoxetine
saquinavir	chloramphenicol	starfruit
telithromycin	ciprofloxacin	voriconazole

CYP3A4 Inducers

Aminoglutethimide	Nevirapine	Phenytoin	Rifapentine
Carbamazepine	Oxcarbazepine	Primidone	St. John's wort (3)
Fosphenytoin	Pentobarbital	Rifabutin	
Nafcillin	Phenobarbital	Rifampin	

When Study Agent is co-administered with drugs classified as "substrates," the plasma concentration of the substrate is high. When Study Agent is co-administered with drugs classified as 'inhibitors,' plasma concentrations of the Study Agent will be high. When Study Agent is co-administered with drugs classified as "inducers," the plasma concentration of the Study Agent will be low.

In general, drug interactions occur significantly between substrates and either inhibitors or inducers of the same enzymes usually classified as "strong" substrates, inhibitors, or inducers.

Note: Adapted from Cytochrome P450 Enzymes: Substrates, Inhibitors, and Inducers. In: Lacy CF, Armstrong LL, Goldman MP, Lance LL eds. Drug Information Handbook 15TH ed. Hudson, OH; LexiComp Inc. 2007: 1899-1912.

Only major substrates and effective inducers are listed.

Additional information for drug interactions with cytochrome P450 isoenzymes can be found at http://medicine.iupui.edu/flockhart/.

(1) Investigator's Brochure, MK-2206. Merck.. January 2009.

(2) Malhotra et al. (2001). Clin Pharmacol Ther. 69:14-23.

(3) Mathijssen et al. (2002). J Natl Cancer Inst. 94:1247-1249.

Frye et al. (2004). Clin Pharmacol Ther. 76:323-329.

Updated on May 1, 2007

18.7 Appendix G: INFORMATION ON POSSIBLE INTERACTIONS WITH OTHER AGENTS FOR PATIENTS AND THEIR CAREGIVERS AND NON-STUDY HEALTH CARE TEAM

The patient _____ is enrolled on a clinical trial using the experimental agent _____. 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.

_____ interacts with many drugs that are processed by your liver. Because of this, it is very important to tell your study doctors about all of your medicine before you start this study. It is also very important to tell them if you stop taking any regular medicine, or if you start taking a new medicine while you take part in this study. When you talk about your medicine with your study doctor, include medicine you buy without a prescription at the drug store (over-the counter remedy), or anything that you buy from the health food store or grocery store (herbal supplement).

Many health care prescribers can write prescriptions. You must also tell your other prescribers (doctors, physicians' assistants or nurse practitioners) that you are taking part in a clinical trial. Bring this paper with you. These are the things that you and they need to know:

- _____ is metabolized (converted in the body) by a liver enzyme called CYP3A4. _____ must be used very carefully with other medicines that need this liver enzyme to be effective or to be cleared from your system.
- You and healthcare providers who prescribe drugs for you must be careful about adding or removing any drug in this category.
- Before you start the study, your study doctor will work with your regular prescriber to switch any medicines that are considered "strong inducers/inhibitors or substrates of CYP3A4."
- Your regular prescribers should look at this web site http://medicine.iupui.edu/clinpharm/ddis/table.asp to see if any medicine they want to prescribe is on a list of drugs to avoid.
- Please be very careful! Over-the-counter drugs have a brand name on the label—it's usually big and catches your eye. They also have a generic name—it's usually small and printed on the ingredient list. Find the generic name (your pharmacist can help) and look at the table on the back of this page. Be careful.
- You should not take St. John's wort or grapefruit juice with _____.
- You should not receive steroids unless they are absolutely necessary; tell your study doctor if you are taking, have a prescription for, or have been given steroids.
- You should not take drugs that affect your heart rhythm. Tell your study doctor if you are taking, have a prescription for, or have been given such heart medications.
- Other medicines can be a problem with your study drugs.

- You should check with your doctor or pharmacist whenever you need to use an over-the-counter medicine or herbal supplement.
- Your regular prescriber should check a medical reference or call your study doctor before prescribing any new medicine for you. Your study doctor's name is _____ and he or she can be contacted at _____.

18.8 Appendix H: Hypertension management tools for patients in sunitinib

Oral Antihypertensive Medications

Agents in bold characters are suggested as optimal choices to avoid or minimize potential drug-interactions with sunitinib through CYP450.

Agent class	Agent	Initial dose	Intermediate dose	Maximum dose	Hepatic metabolism
Dihydro-pyridine Calcium-Channel Blockers (DHP CCB)	**nifedipine XL**	**30 mg daily**	**60 mg daily**	**90 mg daily**	**CYP 3A4 substrate**
	amlodipine	**2.5 mg daily**	**5 mg daily**	**10 mg daily**	**CYP 3A4 substrate**
	felodipine	2.5 mg daily	5 mg daily	10 mg daily	CYP 3A4 substrate and inhibitor
Selective β Blockers (BB)	metoprolol	25 mg twice daily	50 mg twice daily	100 mg twice daily	CYP 2D6 substrate
	atenolol	**25 mg daily**	**50 mg daily**	**100 mg daily**	**No**
	acebutolol	100 mg twice daily	200-300 mg twice daily	400 mg twice daily	Yes (CYP450 unknown)
	bisoprolol	2.5 mg daily	5-10 mg daily	20 mg daily	Yes (CYP450 unknown)
Angiotensin Converting Enzyme Inhibitors (ACEIs)	captopril	12.5 mg 3x daily	25 mg 3x daily	50 mg 3x daily	CYP 2D6 substrate
	enalapril	5 mg daily	10-20 mg daily	40 mg daily	CYP 3A4 substrate
	ramipril	2.5 mg daily	5 mg daily	10 mg daily	Yes (CYP450 unknown)
	lisinopril	**5 mg daily**	**10-20 mg daily**	**40 mg daily**	**No**
	fosinopril	10 mg daily	20 mg daily	40 mg daily	Yes (CYP450 unknown)
	Rarely used: **perindopril**	**4 mg daily**	**none**	**8 mg daily**	**Yes, but not CYP450**
	Rarely used: **quinapril**	**10 mg daily**	**20 mg daily**	**40 mg daily**	**No**
Angiotensin II Receptor Blockers (ARBs)	losartan	25 mg daily	50 mg daily	100 mg daily	CYP 3A4 substrate
	candesartan	4 mg daily	8-16 mg daily	32 mg daily	CYP 2C9 substrate
	irbesartan	75 mg daily	150 mg daily	300 mg daily	CYP 2C9 substrate
	telmisartan	**40 mg daily**	**none**	**80 mg daily**	**Yes, but not CYP450**

	valsartan	**80 mg daily**	**none**	**160 mg daily**	**Yes, but not CYP450**
α and β Blocker	labetolol	100 mg twice daily	200 mg twice daily	400 mg twice daily	CYP 2D6 substrate and inhibitor

Collection/Recording of Blood Pressure Information

1.0 General Guidelines

1.1 Frequency of monitoring. Blood pressure (BP) should be monitored weekly during the first cycle of sunitinib therapy, then at least every 2 weeks for the duration of treatment. More frequent monitoring may be considered on a study by study basis, particularly during the first two cycles of sunitinib therapy.

1.2 Data recording. All required data should be recorded in the appropriate CRF or on the patient's blood pressure monitoring diary, as appropriate. The following data are required at baseline and at each subsequent assessment:

- Assessment date and time
- Pulse
- Systolic and diastolic BP (2 readings/assessment taken 5 minutes apart while patient sitting)

1.3 Risk factors for hypertension (assess and record data in baseline history/physical CRF)

- Diabetes (type 1 or type 2)
- Renal disease (specify on CRF)
- Endocrine condition associated with HTN (specify on CRF)
- Use of steroids or NSAIDs (specify all concomitant meds)
- Underlying cardiovascular condition – specify (i.e., ischemic heart disease)

2.0 Baseline data collection (at study arm entry)

2.1 All patients

- Current BP
- Proteinuria, if present

2.2 Patients with preexisting hypertension (i.e., those for whom "hypertension" is entered as a concomitant condition at study entry, or those who are currently receiving therapy with antihypertensive medication) – also record:

- Date of HTN diagnosis (original)
- Type HTN (essential or secondary)
- CTCAE grade of HTN (at time of study entry)

- Trade name, drug class*, dose, dose frequency, start/stop dates/ongoing of the following:
    - Antihypertensive agents taken at study entry
    - Antihypertensive agents taken in past (e.g., discontinued for toxicity, lack of efficacy)

3.0 Follow up BP data collection (during study)

3.1 All patients (at each clinic visit)

- Current BP
- Proteinuria, if present

3.2 Patients with treatment-emergent hypertension [defined as BP increase of >20 mmHg (diastolic) OR systolic BP >139 OR diastolic BP > 90 (if previously normal or grade 1 per CTCAE v4) – record at time of hypertension diagnosis and at all subsequent clinic visits:

- BP changes from baseline (or from previous assessment) (specify CTCAE grade changes)
- Hypertension-related symptoms as reported by patient (e.g., headache)
- Other relevant changes associated with development of hypertension (e.g., ECG abnormalities)
- Trade name, drug class*, dose, dose frequency, start/stop dates/ongoing of currently prescribed antihypertensive agents

3.3 Patients with pre-existing hypertension at study entry – record at each clinic visit

- BP changes from previous clinic visit (specify CTCAE grade changes)
- Hypertension-related symptoms reported by patient (e.g., headache)
- Other relevant changes associated with development of hypertension (e.g., ECG abnormalities)
- Changes in antihypertensive medications since last assessment (e.g., dose change, add/discontinue drug)

Classes of antihypertensive drugs include ACE inhibitors, calcium channel blockers, alpha blockers, beta blockers, diuretics, angiotension II receptor antagonists.

18.9 Appendix I: CTEP MULTICENTER GUIDELINES

If an institution wishes to collaborate with other participating institutions in performing a CTEP sponsored research protocol, then the following guidelines must be followed.

Responsibility of the Protocol Chair

- The Protocol Chair will be the single liaison with the CTEP Protocol and Information Office (PIO). The Protocol Chair is responsible for the coordination, development, submission, and approval of the protocol as well as its subsequent amendments. The protocol must not be rewritten or modified by anyone other than the Protocol Chair. There will be only one version of the protocol, and each participating institution will use that document. The Protocol Chair is responsible for assuring that all participating institutions are using the correct version of the protocol.
- The Protocol Chair is responsible for the overall conduct of the study at all participating institutions and for monitoring its progress. All reporting requirements to CTEP are the responsibility of the Protocol Chair.
- The Protocol Chair is responsible for the timely review of Adverse Events (AE) to assure safety of the patients.
- The Protocol Chair will be responsible for the review of and timely submission of data for study analysis.

Responsibilities of the Coordinating Center

- Each participating institution will have an appropriate assurance on file with the Office for Human Research Protection (OHRP), NIH. The Coordinating Center is responsible for assuring that each participating institution has an OHRP assurance and must maintain copies of IRB approvals from each participating site.
- Prior to the activation of the protocol at each participating institution, an OHRP form 310 (documentation of IRB approval) must be submitted to the CTEP PIO.
- The Coordinating Center is responsible for central patient registration. The Coordinating Center is responsible for assuring that IRB approval has been obtained at each participating site prior to the first patient registration from that site.
- The Coordinating Center is responsible for the preparation of all submitted data for review by the Protocol Chair.

The Coordinating Center will maintain documentation of AE reports. There are two options for AE reporting: (1) participating institutions may report directly to CTEP with a copy to the Coordinating Center, or (2) participating institutions report to the Coordinating Center who in turn report to CTEP. The Coordinating Center will submit AE reports to the Protocol Chair for timely review.

Audits may be accomplished in one of two ways: (1) source documents and research records for selected patients are brought from participating sites to the Coordinating Center for audit, or (2) selected patient records may be audited on-site at participating sites. If the NCI chooses to have an audit at the Coordinating Center, then the Coordinating Center is responsible for having all source documents, research records, all IRB approval documents, NCI Drug Accountability Record forms, patient registration lists, response assessments scans, x-rays, etc. available for the audit.

Inclusion of Multicenter Guidelines in the Protocol

- The protocol must include the following minimum information:
- The title page must include the name and address of each participating institution and the name, telephone number and e-mail address of the responsible investigator at each participating institution.
- The Coordinating Center must be designated on the title page.
- Central registration of patients is required. The procedures for registration must be stated in the protocol.
- Data collection forms should be of a common format. Sample forms should be submitted with the protocol. The frequency and timing of data submission forms to the Coordinating Center should be stated.
- Describe how AEs will be reported from the participating institutions, either directly to CTEP or through the Coordinating Center.
- Describe how Safety Reports and Action Letters from CTEP will be distributed to participating institutions.

Agent Ordering

- Except in very unusual circumstances, each participating institution will order DCTD-supplied agents directly from CTEP. Agents may be ordered by a participating site only after the initial IRB approval for the site has been forwarded by the Coordinating Center to the CTEP PIO.

18.10 Appendix J: Molecular Profiling Follow-Up Worksheet

Patient Name:______________________________

Circle: Living or Deceased

If deceased, DOD:______________

Further Chemotherapy/Biotherapy Treatment(s)

Date of follow up:______________ **Completed By:**______________

Drug Name:______________________________

Dates of administration: **Start:**__________ **Stop:**__________

Reason Treatment ended: Progression / Other

Entered into database by:______________ **Date:**______________

Date of follow up:______________ **Completed By:**______________

Drug Name:______________________________

Dates of administration: **Start:**__________ **Stop:**__________

Reason Treatment ended: Progression / Other

Entered into database by:______________ **Date:**______________

Date of follow up:______________ **Completed By:**______________

Drug Name:______________________________

Dates of administration: **Start:**__________ **Stop:**__________

Reason Treatment ended: Progression / Other

Entered into database by:______________ **Date:**______________

Date of follow up:______________ **Completed By:**______________

Drug Name:______________________________

Dates of administration: **Start:**__________ **Stop:**__________

Reason Treatment ended: Progression / Other

Entered into database by:______________ **Date:**______________

18.11 Appendix K: Tobbacco Use History Questionnaire

TOBACCO USE HISTORY

Patient Name_____ Date _____

Have you ever used any tobacco products? _____

If yes, did you smoke cigarettes?_____

What is the average number of cigarettes you smoke(d) each day?_____

Did you smoke cigars? _____ How many cigars did you smoke each day?_____

Did you smoke a pipe? _____ How many pouches did you use each day?_____

Did you chew tobacco? ____ How many pouches did you use each day?_____

Did you use snuff? _____ How many cans did you use each day? _____

How old were you when you started?_____

At what age did you stop? _____

Do you currently use tobacco? _____

Have you been exposed to smoke from other people? _____

If yes, who smoked in your presence?

____spouse ____parent ____sibling ____other

____ number of cigarettes that other people smoked in your presence on an average day

____ number of years you were exposed to other people smoking in your presence

Patient Signature: _____ Date: _____

Provider Signature: _____ Date: _____

18.12 Appendix L: Acquisition, Storage, Interpretation and Reporting of Low-Dose CT Scans for Enrolled First-Degree Relatives

Low-dose computed tomography acquisition parameters (adapted from NCCN Guidelines for Lung Cancer Screening, version 1.2012)		
Acquisition	Small patient (BMI ≤30)	Large patient (BMI >30)
Total radiation exposure	≤ 3 mSv	≤ 5 mSv
kVp	100-120	120
mAs	≤ 40	≤ 60
All patients		
Gantry rotation speed	≤ 0.5	
Detector collimation	≤ 1.5 mm	
Slice width	≤ slice width; 50% overlap preferred for 3D and CAD applications	
Scan acquisition time	≤ 10 seconds (single breath hold)	
Breathing	Maximum inspiration	
Contrast	No oral or intravenous contrast	
Ct scanner detectors	≥ 16	
Abbreviations: CAD: computed aided diagnostics; BMI: body mass index		