UCSF Helen Diller Family Comprehensive Cancer Center

NCT# 01687673

Phase II Trial of the Combination of Temsirolimus and Sorafenib in Advanced Hepatocellular Carcinoma

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Protocol Signature Page

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1. I agree to follow this protocol version as approved by the UCSF Protocol Review Committee (PRC), Committee on Human Research (CHR), and Data Safety Monitoring Committee (DSMC).
2. I will conduct the study in accordance with applicable CHR requirements, Federal regulations, and state and local laws to maintain the protection of the rights and welfare of study participants.
3. I certify that I, and the study staff, have received the requisite training to conduct this research protocol.
4. I have read and understand the information in the Investigators' Brochure (or Manufacturer's Brochure) regarding the risks and potential benefits. I agree to conduct the protocol in accordance with Good Clinical Practices (ICH-GCP), the applicable ethical principles, the Statement of Investigator (Form FDA 1572), and with local regulatory requirements. In accordance with the FDA Modernization Act, I will ensure the registration of the trial on the www.clinicaltrials.gov website.
5. I agree to maintain adequate and accurate records in accordance with CHR policies, Federal, state and local laws and regulations.

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_____
Printed Name

_____ _____
Signature Date

Participating Sites

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Principal Investigator **Site**

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Printed Name

_____ _____
Signature Date

Abstract

Title	Phase II Trial of the Combination of Temsirolimus and Sorafenib in Advanced Hepatocellular Carcinoma
Patient population	Histologically-confirmed, advanced, treatment-naïve HCC with Child-Pugh A or B7 liver dysfunction
Rationale for Study	To determine efficacy of and explore biomarkers for response to combination of temsirolimus plus sorafenib in advanced HCC
Primary Objective	To determine the median time to progression (TTP) in patients with advanced HCC treated with combination of temsirolimus plus sorafenib
Secondary Objectives	1. To determine response rate (RR) using RECIST 1.1 2. To measure median progression-free survival (PFS) 3. To measure median overall survival (OS) 4. To measure time to treatment failure (TTF) 5. To measure change in AFP tumor marker and examine for association between change in AFP and TTP and best response 6. To characterize toxicity and tolerability of the combination of temsirolimus plus sorafenib 7. To describe TTP, PFS, OS, response rate, AFP response, and toxicity and tolerability in subsets defined by HBV, HCV, Asian, and non-Asian status
Study Design	Single-arm, one-stage, open-label phase II trial to evaluate the efficacy of the combination of temsirolimus plus sorafenib in first line therapy for patients with histologically-confirmed advanced HCC.
Number of patients	Target sample size is 25 patients, expected maximum enrollment is 28 patients.
Duration of Therapy	Individual patient treatment will continue indefinitely from the time of enrollment, until unacceptable toxicity, unmanageable toxicity despite optimal supportive care and dose modifications, clinical disease progression or progression per RECIST 1.1, withdrawal of consent by the patient, or the decision to discontinue treatment by the treating physician due to changes in the patient's condition or other factors which may influence safety or compliance.
Duration of Follow up	After treatment discontinuation, follow up for overall survival monitoring will be performed for up to 5 years or until death, whichever comes first.
Duration of study	The study will reach completion approximately 24 months from the time the study opens to accrual. The study will end 5 years after the final patient discontinues treatment or after all patients are deceased, whichever comes first.

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Study Drugs	Temsirolimus: 10 mg weekly will be administered intravenously over 30 to 60 minutes using an infusion pump starting on Cycle 1, Day 1 of study enrollment. Sorafenib: 200 mg tablet twice daily starting on Cycle 1, Day 1 of study enrollment after completion of temsirolimus infusion.
Safety Assessments	Analyses will be performed for all patients having received at least one dose of study drug. The study will use the NCI (CTCAE) Version 4.0.
Efficacy Assessments	Primary Endpoint Median TTP will be calculated in months from date of first dose of protocol therapy to date of removal from study for progression; Kaplan-Meier methods will be used to summarize the primary endpoint (median TTP). Secondary Endpoints Response rate (RR) will be measured by RECIST version 1.1; measurements will be presented descriptively for the study cohort. Median PFS will be calculated in months from date of first dose of protocol therapy to date of documented disease progression or death from any cause. Kaplan-Meier methods will be used to summarize time-to-event outcomes including the primary endpoint (median TTP) and secondary endpoints (PFS, TTF, OS). Median OS for all enrolled patients who receive at least one dose of protocol therapy will be calculated from date of first dose of protocol therapy until date of death, using chart review and/or follow up phone calls to determine date of death in patients after removal from study. TTF will be measured from date of first dose of protocol therapy to date of study discontinuation for progression, death, or toxicity. In patients with baseline AFP ≥ 20 ng/mL, AFP response will be measured by the percent change from baseline value to the value at the time of best AFP response. The change in AFP will also be examined for association with absolute TTP and best response.

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Efficacy Assessments (continued)	<u>Exploratory Endpoints</u> CTC will be enumerated in peripheral blood at baseline and after 1 and 2 cycles of therapy to evaluate for any relationship between baseline levels and change in levels with median TTP and/or OS; baseline level of CTC will be compared to proportion with TTP at 6 months to determine if there is an association using Fisher's Exact tests. Change in CTC levels (increase or decrease) after 1 and 2 cycles will be compared to TTP to determine if there is an association using Fisher's Exact tests. In all patients with active or chronic HBV infection, changes in HBV DNA quantitative by PCR will be characterized using descriptive statistics to calculate the proportion of patients with an increase or reactivation in viral load while on protocol therapy.
Unique Aspects of this Study	This Phase II trial is being developed following completion of a Phase I study of the combination of temsirolimus and sorafenib in 25 first-line therapy patients with advanced hepatocellular carcinoma (December 2009 through April 2012). The MTD and recommended phase II dose (RP2D) of the combination was temsirolimus 10 mg IV weekly plus sorafenib 200 mg PO BID.

List of Abbreviations

AE	adverse event
ALP	alkaline phosphatase
ALT	alanine aminotransferase
ANC	absolute neutrophil count
AST	aspartate aminotransferase
ATC	Anatomical Therapeutic Chemical (Classification System)
AUC	area under the curve
BUN	blood urea nitrogen
CBC	complete blood cell (count)
CHR	Committee on Human Research (UCSF IRB)
CR	complete response
CRC	Clinical Research Coordinator
CRF	case report form
CSF	cerebral spinal fluid
CT	computerized tomography
CTCEA	Common Terminology Criteria for Adverse Events
CTEP	Cancer Therapy Evaluation Program
CTMS	Clinical Trial Management System
DFS	disease-free survival
DLT	dose limiting toxicity
DSMC	Data and Safety Monitoring Committee
DSMP	Data and Safety Monitoring Plan
ECOG	Eastern Cooperative Oncology Group
FCBP	female of childbearing potential
FDA	Food and Drug Administration
GCP	Good Clinical Practice
HBeAg	Hepatitis B “e” antigen
HBV	Hepatitis B virus
HCC	Hepatocellular carcinoma
HCT	Hematocrit
HCV	Hepatitis C virus
HDFCCC	Helen Diller Family Comprehensive Cancer Center
HGB	Hemoglobin
HIV	human immunodeficiency virus
ICH	International Conference on Harmonization

List of Abbreviations

IND	investigational new drug application
IP	investigational product
IRB	Institutional Review Board
iwCLL	International Workshop on Chronic Lymphocytic Leukemia
IV	Intravenous
LDH	lactate dehydrogenase
LFT	liver function test
MedDRA	Medical Dictionary for Regulatory Activities
MRI	magnetic resonance imaging
MTD	maximum tolerated dose
NCI	National Cancer Institute
NHL	non-Hodgkin's lymphoma
ORR	overall response rate
PD	disease progression
PK	Pharmacokinetics
PO	*Per os* (by mouth, orally)
PR	partial response
PRC	Protocol Review Committee (UCSF)
QOL	Quality of Life
RBC	red blood cell (count)
SD	stable disease
SD	standard deviation
SGOT	serum glutamic oxaloacetic transaminase
SGPT	serum glutamic pyruvic transaminase
ULN	upper limit of normal
WBC	white blood cell (count)

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1. Introduction

1.1 Background on Indication

Hepatocellular carcinoma (HCC) is the third leading cause of cancer death worldwide with an incidence of over 600,000 new cases and almost as many deaths annually. (Parkin, Bray et al. 2005) Advanced stages of disease at diagnosis often preclude curative treatments, and the overall prognosis of patients diagnosed with advanced HCC remains dismal with median survival of approximately 8 months.(Llovet, Burroughs et al. 2003; El-Serag 2007; Llovet, Ricci et al. 2008) Historically, systemic therapies for advanced HCC have not been associated with a survival benefit, largely due to compromised hepatic function from underlying liver disease as well as intrinsic tumor chemoresistance.(Chenivesse, Franco et al. 1993; Soini, Virkajarvi et al. 1996; Jiang, Lu et al. 1997; Yeo, Mok et al. 2005; Lopez, Villanueva et al. 2006) In 2008, publication of the Sorafenib Hepatocellular Carcinoma Assessment Randomized Protocol (SHARP) phase III trial demonstrated a significant improvement in overall survival (OS) in patients with advanced HCC treated with the biologic agent, sorafenib.(Llovet, Ricci et al. 2008) The SHARP trial will be discussed in detail below. Despite improvements in outcome with sorafenib, however, the median OS for patients with advanced HCC remains less than a year.(Llovet, Ricci et al. 2008) Multiple recent randomized phase 3 trials of other anti-angiogenic multikinase inhibitors in HCC, including a first-line trial of sunitinib versus sorafenib and a second-line trial of brivanib versus placebo, have failed to show benefit, underscoring the challenges of toxicity, comorbidity, and chemoresistance in this notoriously treatment-refractory disease.(Cheng 2011; Llovet 2012) New active drugs and combinations are urgently needed.

1.2 Background on the Compounds

1.2.1 Sorafenib

Sorafenib is a small molecule bi-aryl urea with multikinase inhibitor activity. A primary target is the serine-threonine kinase, Raf-1, as well as mutant RET, mutant FLT-3, and wild-type and mutant c-KIT.(Bayer 2011) Sorafenib also has antiangiogenic activity, inhibiting receptor tyrosine kinases including vascular endothelial growth factor (VEGF) receptors 2 and 3 and the platelet derived growth factor receptor-β (PDGFRβ).(Wilhelm, Carter et al. 2004; Liu, Cao et al. 2006; Chang, Adnane et al. 2007) Preclinical studies in HCC show that Raf-1 kinase signaling along with prolific tumor angiogenesis are common features, providing a molecular rationale for the efficacy of this agent.(Huynh, Nguyen et al. 2003; Semela and Dufour 2004; Calvisi, Ladu et al. 2006; Liu, Cao et al. 2006; Villanueva, Newell et al. 2007) In preclinical models of HCC including athyrmic mouse xenografts, sorafenib treatment produces both anti-tumor and anti-angiogenic effects.(Bayer 2011)

The metabolism of sorafenib is predominantly hepatic via oxidation by CYP3A4 and glucuronidation by UGT1A9.(Bayer 2011) Sorafenib is a moderate to strong inhibitor of CYP2C8, 2B6, 2C9, 2C19, 2D6, and 3A4 as well as UGT1A1 and 1A9. In cultured hepatocytes, sorafenib does not induce CYP1A2 or 3A4. Excretion is predominantly through biliary/fecal route. Bioavailability is moderate to high in animal models. Terminal half-life from plasma is 4 hours in dogs, 9 hours in rats, and 6 hours in mice. There is high plasma protein binding of 99.5%. The sorafenib metabolites M2 and M5 also potently inhibit VEGFR2, PDGFRβ, and

FLT-3, though in xenograft models, M2 has lower efficacy than the parent compound. In preclinical models, sorafenib can be safely combined with multiple conventional cytotoxic agents including gemcitabine and cisplatin.

In human studies, there was evidence of accumulation up to 7-fold upon multiple dosing without further increase after 7 days of multiple dosing. The mean terminal elimination half-life in humans is 25 to 48 hours. It should be administered fasting or with a low- or moderate-fat meal, as high-fat meals decrease bioavailability. In humans, 19% of dose is excreted in urine and 77% in feces. M2 is the principle plasma metabolite. There is high inter-patient PK variability not explained by gender, age, weight, or hepatic function. Prior phase I studies of sorafenib have shown significant interpatient pharmacokinetic (PK) variability without clear relationship to toxicity.(Strumberg, Richly et al. 2005; Strumberg, Clark et al. 2007) The exposure values and PK are similar in patients with Child-Pugh A and B hepatic dysfunction. In a phase I study studying sorafenib in patients with organ dysfunction, there was no clear effect of renal or hepatic dysfunction on sorafenib clearance by PK measures, though toxicity was increased.(Miller, Murry et al. 2009) No dose adjustment is required for patients with mild to severe renal impairment not requiring dialysis. The mean AUC of sorafenib was 30% lower in Asians than Caucasians but does not appear to correlate with toxicity or efficacy. There was minimal impact on cardiovascular parameters in humans though there is a small to minimal mean prolongation of QT/QTc interval. Human metabolism is predominantly hepatic through CYP3A4-mediated oxidation and UGT1A9-mediated glucuronidation. It may be co-administered with CYP3A4 substrates and inhibitors without expected increase in exposure, though inducers such as rifampin may reduce sorafenib AUC. Alteration of GI flora by antibiotics may also reduce exposure. Other agents and classes with potential for drug interactions with sorafenib are reviewed in *Section 6.7* and *Appendices 6-9 Prohibited Medications*.

Additional information on sorafenib is provided in *Section 4 Study Drugs* as well as in the Investigator's Brochure. (Bayer 2011)

1.2.1.1 Phase I and II Clinical Studies of Sorafenib in HCC

In the phase I setting, sorafenib demonstrated an acceptable safety profile at a dose of 400 mg orally twice daily in patients with advanced solid tumors; there was a partial response in a patient with HCC.(Strumberg, Richly et al. 2005) A multicenter phase II trial of sorafenib in advanced HCC was published by Abou-Alfa et al in 2006.(Abou-Alfa, Schwartz et al. 2006) In this study, 137 patients with unresectable HCC were treated with sorafenib 400 mg twice daily. Almost half of patients (48%) were positive for hepatitis C virus (HCV) infection, while 17% had hepatitis B virus (HBV). The majority demonstrated preserved liver function, with a score of Child-Pugh class A in 72% and Child-Pugh class B in 28%. There were no significant differences in sorafenib pharmacokinetic parameters between Child-Pugh class A and B patients. Overall, the toxicity profile of sorafenib was acceptable, with the most common Grade 3 or 4 toxicities being fatigue, diarrhea, and hand-foot skin reaction in 9.5%, 8.0%, and 5.1% of patients, respectively. There were no patients with a complete response (CR), 11 (8%) with partial and minor responses, and 46 (33.6%) with stable disease for at least 16 weeks. Many tumors seemed to have necrosis, however, including some with apparent size increase by conventional imaging. Survival parameters were comparable to those of the best published combinations of systemic chemotherapy in advanced HCC.(Yeo, Mok et al. 2005; Abou-Alfa, Schwartz et al. 2006; Lopez, Villanueva et al. 2006) Despite no significant difference in pharmacokinetic profiles based upon liver function, subgroup analysis of the 28% of patients with Child-Pugh class B cirrhosis in this phase II study showed higher rates of hyperbilirubinemia and encephalopathy developing on study by comparison with patients with Child-Pugh class A.(Abou-Alfa, Amadori et al. 2011) Patients with Child-Pugh class B liver function also demonstrated a shorter median OS of 3.2 months by comparison to 9.5 months for Child-Pugh class A patients.

1.2.1.2 Phase III Clinical Studies of Sorafenib in HCC

Following upon this phase II study of sorafenib in HCC, the phase III multicenter SHARP trial enrolled 602 patients with advanced HCC and preserved liver function from 121 sites in Europe, North America, South America, and Australasia. The patient population was comprised of approximately 30% with HCV infection, 20% with HBV infection, and 25% with alcoholic liver disease. Nearly all patients had Child-Pugh class A cirrhosis, with Eastern Cooperative Oncology Group performance status score of 0 or 1. Patients were randomly assigned to receive sorafenib at a starting dose of 400 mg twice daily versus placebo. The primary end points were overall survival (OS) and time to symptomatic progression, and patients could remain on treatment until they experienced disease progression both radiographically and symptomatically. At the second planned interim analysis, OS was significantly longer in the sorafenib group compared with placebo, with OS of 10.7 months versus 7.9 months, respectively, and hazard ratio (HR) for sorafenib of 0.69 ($P < 0.001$). There was no difference in time to symptomatic progression, though median time to radiographic progression (TTP) was significantly longer in the sorafenib group (5.5 vs 2.8 months, $p < 0.001$). Seven partial responses by Response Evaluation Criteria in Solid Tumors (RECIST) occurred in the sorafenib-treated arm (2%). Disease control rate (DCR) (a composite of complete response, partial response, and stable disease) was significantly higher in the sorafenib group (43% v 32%; $p < 0.002$).

Subgroup analysis from SHARP suggested that patients with HCV have similar safety profile and clinical benefit as the entire study population, with a trend towards greater survival benefit with median OS 14.0 months for patients with HCV by comparison with 10.7 months for the overall population.(Bolondi L. 2008) Patients with alcohol-related HCC and with macroscopic vascular invasion and/or extrahepatic spread also demonstrated survival benefit with sorafenib and similar toxicity profile as the overall study population by subgroup analyses from SHARP.(Craxi A. 2008; Sherman M 2008) Subgroup analysis for patients with HBV in the SHARP study has not been presented to date.

Another randomized, placebo-controlled phase III study comparing sorafenib to placebo was conducted in 226 Asian patients with unresectable HCC, Child-Pugh class A cirrhosis, and greater than 70% with underlying HBV infection.(Cheng, Kang et al. 2009) Survival was significantly prolonged with the use of sorafenib, with median OS 6.5 months in patients treated with sorafenib by comparison with 4.2 months for placebo (HR 0.68, $p = 0.014$). Median TTP was 2.8 months in sorafenib group compared to 1.4 months in the placebo group (HR 0.57, $p = 0.0005$). Though sorafenib produced clear benefit, the outcomes in this Asian study were much poorer than those of SHARP. Potential explanations include possible differences in numbers of prior therapies, assessment of performance status, exposure to other factors influencing hepatocarcinogenesis, or etiology of liver disease between the Asian population and the SHARP population.(Kelley and Venook 2008)

1.2.1.3 Safety of Sorafenib in HCC

In the SHARP study, treatment-related adverse events occurred in 80% of patients in the sorafenib group by comparison with 52% in the placebo group. Grade 3 toxicities included diarrhea (8%), hand-foot skin reaction (8%), hypertension (2%), and abdominal pain (2%) in the treatment group. Grade 3 or 4 hypophosphatemia (4%) and thrombocytopenia (4%) were also more common in patients treated with sorafenib.(Llovet, Ricci et al. 2008) In the Asian randomized, placebo-controlled phase III study, the most common Grade 3 and 4 toxicities were hand-foot skin reaction (10.1%), diarrhea (6%), hyperbilirubinemia (3.4%), and fatigue (3.4%) with the use of sorafenib.(Cheng, Kang et al. 2009) As discussed above, subgroup analysis from the phase II study of sorafenib in HCC of the 28% of patients with Child-Pugh class B cirrhosis showed higher rates of hyperbilirubinemia and encephalopathy developing on study by comparison with patients with Child-Pugh class A.(Abou-Alfa, Amadori et al. 2011) A

retrospective single-institution study of 48 patients showed significantly decreased tolerability of sorafenib among Asian-American patients compared to non-Asian patients, with only 3% tolerating full-dose sorafenib of 400 mg twice daily compared to 40% for non-Asians (P < 0.01).(Barrera 2009) Safety data for sorafenib are reviewed in greater detail below in Section 4.1.2 as well as in the FDA Prescribing Information.(Bayer 2011)

1.2.1.4 Mammalian Target of Rapamycin (mTOR) Inhibition in Malignancy

Another signaling pathway involved in hepatocarcinogenesis is the phosphatase and tensin homolog (PTEN)/phosphatidylinositol-3'kinase (PI-3'K)/AKT pathway which activates the mammalian target of rapamycin (mTOR) kinase, in turn triggering multiple downstream cell growth, survival, and angiogenesis signals.(Dancey 2006; Hopfner, Schuppan et al. 2008; Llovet and Bruix 2008) Dysregulated phosphorylation and activation of mTOR signaling may occur due to loss of function of the PTEN tumor suppressor gene, constitutive activation of PI-3'K, or activation of AKT by aberrant upstream growth factor receptor signaling.(Llovet and Bruix 2008) Activated mTOR forms complexes with other proteins, including regulatory associated protein of mTOR (Raptor) and Rictor.(Dancey 2006) The mTOR-Raptor complex in turn phosphorylates protein 70 S6 kinase (p70S6K) as well as eukaryotic initiation factor 4E (eIF-4E) binding protein-1 (4E-BP1). P70S6K and 4E-BP1 regulate translation of a host of proteins, including several proteins involved in cell proliferation. Signaling through mTOR also stimulates angiogenesis.(Dancey 2006; Semela, Piguet et al. 2007; Hopfner, Schuppan et al. 2008; Villanueva, Chiang et al. 2008) Activation of mTOR may induce endothelial cell proliferation as well as increase levels of hypoxia inducible factor (HIF)-1α and HIF-2α, potentially via p70S6K-mediated translation versus decreased oxygen-dependent degradation.(Zhong, Chiles et al. 2000; Hudson, Liu et al. 2002; Thomas, Tran et al. 2006) HIFs induce angiogenesis in response to cellular hypoxia by transcriptional activation of target genes including VEGF.(Kaelin 2005; Del Bufalo, Ciuffreda et al. 2006; Thomas, Tran et al. 2006)

The mTOR inhibitor, sirolimus, is a macrocyclic lactone rapamycin produced by the soil bacterium, Streptomyces hygroscopicus. Sirolimus demonstrates fungicidal, immunosuppressive, and antiproliferative properties and is widely used as an immunosuppressant in transplant patients to prevent allograft rejection.(Douros and Suffness 1981; Dancey 2006) Temsirolimus, everolimus, and deforolimus are derivatives of sirolimus with similar antiproliferative properties in vitro.(Dancey 2006) Inhibitors of mTOR are also under investigation as anti-cancer agents in multiple human malignancies, and two mTOR inhibitors have been labeled by the U.S. FDA for use as anti-cancer agents: Everolimus is indicated in the treatment of metastatic pancreatic neuroendocrine tumors and advanced RCC, and temsirolimus is indicated for the treatment of advanced RCC.

1.2.1.5 Non-Clinical Studies of mTOR Pathway Inhibition in HCC

In vitro, mTOR activation appears exert an oncogenic effect on hepatocellular cell lines. In the non-hepatoma hepatocellular cell line HepaRG, a constitutively active mTOR mutation confers a premalignant phenotype, though mutations in mTOR, PTEN, and PIK3CA are rare in studies of clinical specimens.(Parent, Kolippakkam et al. 2007; Villanueva, Chiang et al. 2008) A significant proportion of resected HCC tumor specimens feature activation of the mTOR pathway, as measured by phosphorylation of one or multiple members of the mTOR cascade including mTOR itself, AKT, IGF-1R, 4EBP-1, p-RPS6, and p70S6K, or by increased levels of total p70S6K expression.(Sahin, Kannangai et al. 2004; Sieghart, Fuereder et al. 2007; Schmitz, Wohlschlaeger et al. 2008; Villanueva, Chiang et al. 2008; Baba, Wohlschlaeger et al. 2009) In a study of 166 liver explants with HCC from orthotopic liver transplant recipients, phosphorylated mTOR (p-mTOR) was identified by immunohistochemistry in 41% of tumors; p-mTOR was not identified in adjacent non-tumor cirrhotic tissue.(Sieghart, Fuereder et al. 2007) Phosphorylated p70S6K was present in 49% of HCC tumors in this study, and phosphorylated 4EBP-1 was present in 39%. There was no apparent correlation, however, between mTOR

activation and disease-free or overall survival. In another study of 101 resected hepatoma specimens, phosphorylated AKT (p-AKT) measured by immunohistochemistry was present with moderate levels in 50.5% and strong levels in 31.4%, by comparison with no staining in normal hepatocellular tissue.(Schmitz, Wohlschlaeger et al. 2008) Tumors with moderate and strong p-AKT expression were associated with a trend toward reduced tumor-specific overall survival (P = 0.059) in this study. Two other large studies of HCC resection specimens detected elevated levels of phosphorylated p70S6K in 24.5% and p-RPS6 in 47.7% of tumors, respectively, using immunohistochemistry; the presence of phosphorylated p70S6K or p-RPS6 was associated with poor clinical outcomes in both studies.(Villanueva, Chiang et al. 2008; Baba, Wohlschlaeger et al. 2009) Phosphorylated AKT, mTOR, and p70S6K have also been detected in HCC-associated sinusoidal endothelial cells by comparison to non-malignant liver tissues.(Li, Tan et al. 2008)

In multiple HCC cell lines, the mTOR inhibitors everolimus and sirolimus have been shown to inhibit proliferation, induce cell cycle arrest, and reduce viability.(Villanueva, Chiang et al. 2008; Wang, Zhou et al. 2008) Treatment with sirolimus has also been shown to reduce VEGF and HIF-1α mRNA expression in vitro.(Wang, Zhou et al. 2008) Similarly, temsirolimus demonstrates an anti-angiogenic effect in endothelial cells as measured by tube formation assays and aortic ring assays.(Semela, Piguet et al. 2007) Xenograft models suggest that mTOR inhibition impairs HCC tumor growth, delays metastatic progression, and prolongs survival.(Semela, Piguet et al. 2007; Huynh, Chow et al. 2008; Villanueva, Chiang et al. 2008; Wang, Zhou et al. 2008) In athymic nude mice injected with Huh-7 HCC cells, treatment with everolimus was associated with a delay in tumor growth as well as decrease in tumor proliferation index as measured by Ki-67 staining by comparison to control mice.(Villanueva, Chiang et al. 2008) In another nude mouse model of the LCI-D20 metastatic HCC cell line, administration of sirolimus impaired tumor growth and metastases.(Wang, Zhou et al. 2008) Tumor microvessel density and circulating levels of VEGF were reduced, suggesting an anti-angiogenic effect. A study in severe combined immunodeficiency (SCID) mice implanted with HCC xenograft tumors demonstrated dose-dependent growth inhibition and decreased tumor proliferative indices; VEGF levels and tumor microvessel density also declined with treatment.(Huynh, Chow et al. 2008) In syngeneic rats implanted with hepatoma cells, treatment with sirolimus also reduced tumor growth and metastases and was associated with reduced tumor microvessel density, again suggesting an anti-angiogenic effect of sirolimus.(Semela, Piguet et al. 2007) Pharmacodynamic studies showed decreased phosphorylation of 4E-BP1 in the treated rats.

1.2.1.6 Clinical Studies of mTOR Pathway Inhibition in HCC

A single-arm phase II trial of sirolimus 20 mg/week in 25 treatment-naïve patients with advanced HCC and Child-Pugh A (N = 17) or B (N = 8) cirrhosis demonstrated an objective response rate of 8%, with 1 patient achieving a complete radiographic response, 1 with a partial response, 8 with stable disease, and 13 with progressive disease at best response; 2 patients had grade 5 toxicity and were unevaluable.(Decaens, Luciani et al. 2012) The median overall survival was 25.4 weeks and median time to radiographic progression was 15.3 weeks. In another small, single-arm, pilot study of 21 patients with HCC and 9 with cholangiocarcinoma in Sweden, treatment with sirolimus to trough levels between 4 and 15 µg/mL resulted in partial response in 1 patient and stable disease in 5 patients with HCC, and stable disease in 3 patients with cholangiocarcinoma.(Rizell, Andersson et al. 2008) A phase I/II trial of everolimus in 28 patients with advanced HCC and 0 to 2 prior systemic treatment regimens showed acceptable tolerability with median PFS of 3.8 months and overall survival of 8.4 months, with PFS of 28.6% at 24 weeks.(Zhu, Abrams et al. 2011) A randomized phase III trial of everolimus versus placebo in advanced HCC patients after failure of sorafenib is ongoing (NCT01035229).

There is emerging evidence that mTOR inhibition confers an anti-tumor effect as well as immunosuppression after orthotopic liver transplantation for HCC. A case report has been published of a liver transplant patient with biopsy-proven recurrent HCC and pulmonary metastases in whom, upon switching immunosuppressive regimen to include the mTOR inhibitor sirolimus, the pulmonary metastases demonstrated a complete radiographic response. (Elsharkawi, Staib et al. 2005) Small studies of liver transplant patients with recurrent HCC after transplant have also suggested a possible antitumor effect when immunosuppression was switched to sirolimus-based immunosuppression.(Kornberg, Kupper et al. 2008; Zhou, Wang et al. 2008) In retrospective analysis of 73 consecutive liver transplant patients at a single center in China, use of sirolimus by comparison with FK506-based immunosuppression was associated with significantly longer overall survival (P = 0.011) and a non-significant trend toward prolonged disease-free survival (P = 0.234).(Zhou, Wang et al. 2008) A meta-analysis of 2950 patients treated with liver transplantation for HCC showed that patients treated with sirolimus-based immunosuppression had lower recurrence risk than those treated with sirolimus-free regimens, with odds ratio of 0.42 (95% confidence interval of 0.21-0.83).(Liang, Wang et al. 2012)

1.2.2 Combination of Sorafenib plus mTOR Inhibition

Combination of molecularly targeted therapies offers the theoretical potential for additive or synergistic inhibition of shared targets as well as targets in parallel pathways which may provide escape mechanisms from single-pathway inhibition. In the case of mTOR inhibitors and Ras/MAPK pathway inhibition, the combination could be addititve or synergistic by blocking a pathway parallel to and/or upstream of mTOR kinase, augmenting anti-angiogenic effects, or blocking the increase in VEGF which has been reported with VEGFR tyrosine kinase inhibition.(Dancey 2006; Deprimo, Bello et al. 2007; Llovet and Bruix 2008; Zhu, Sahani et al. 2009; Martin, Edeline et al. 2012)

1.2.2.1 Preclinical Studies of the Combination of mTOR Inhibition plus Sorafenib in HCC

Xenograft models of HCC have demonstrated additive effects and possible synergy when an mTOR inhibitor is combined with sorafenib or another anti-angiogenic multikinase inhibitor.(Jasinghe, Xie et al. 2008; Wang, Zhou et al. 2008; Huynh, Ngo et al. 2009) In nude mice orthotopically implanted with highly metastatic human HCC tumor cells (LCI-D20), treatment with sirolimus inhibited primary tumor growth as well as lung metastases.(Wang, Zhou et al. 2008) The addition of sorafenib to sirolimus significantly increased anti-tumor effect and augmented suppression of angiogenesis as measured by decreased microvessel density and circulating VEGF levels by comparison with either agent alone. In another study, Huh7 and SK-HEP-1 HCC cells were implanted subcutaneously into SCID mice.(Jasinghe, Xie et al. 2008) Treatment with sirolimus inhibited tumor growth; addition of the multikinase inhibitor ABT-869 produced significantly greater reduction in tumor growth by comparison with controls or with either agent administered alone. A xenograft model of SCID mice implanted with the human HCC cell line 10-0505 and treated with sirolimus, sorafenib, or the combination of both drugs showed that combination therapy induced significant tumor regression as well as growth inhibition by comparison with either drug alone.(Huynh, Ngo et al. 2009) Furthermore, combination therapy with both sorafenib and sirolimus also blocked the upregulation of cyclin B1 and Cdk-2, phosphorylation of c-met, and phosphorylation of mTOR targets that were induced by single agent therapy. VEGFR-2 phopshorylation was likewise inhibited while apoptosis was increased. In rats implanted with Morris Hepatoma cells, treatment with the combination of everolimus plus sorafenib achieved greater tumor reduction than everolimus alone or sequential sorafenib then everolimus, and survival was longest in the combination group ($P < 0.05$). (Piguet, Saar et al. 2011)

1.2.2.2 Clinical Studies of Combination mTOR Inhibition plus Sorafenib

Preliminary results from a phase I clinical trial of the combination of temsirolimus and sorafenib in 24 patients with advanced solid tumors identified partial responses in one patient with non-Hodgkin's lymphoma and two patients with thyroid cancer (one papillary, one medullary); prolonged stable disease was observed in a patient with RCC.(Patnaik A 2007) There were no patients with HCC or documented liver dysfunction in this trial. Starting dosages were temsirolimus 15 mg intravenously weekly plus sorafenib 200 mg twice daily, with escalation to maximum dosages of 25 mg intravenously weekly and 400 mg twice daily, respectively for each drug. Interim results presented at ASCO in 2007 suggested that an intermediate dosage of sorafenib 200 mg twice daily with temsirolimus 25 mg intravenously weekly was tolerable. Significant mucocutaneous toxicity as well as thrombocytopenia were observed with full dosages of both agents. This study has not been published as of April 2012. A phase I study of sorafenib plus temsirolimus in 25 patients with metastatic melanoma showed significant toxicity at higher dose levels, with maximum tolerated dose established as sorafenib 400 mg in the morning and 200 mg every evening plus temsirolimus 25 mg intravenously weekly.(Davies, Fox et al. 2012) Best response was stable disease in 10 patients, and median PFS was 2.1 months. Dose-limiting toxicities were thrombocytopenia, hand-foot syndrome, transaminase elevation, and hypertriglyceridemia. Matching pre-treatment and day 15 tumor biopsies in a subset of 4 patients on this study showed decrease in phospho-S6 but minimal change in phospho-ERK levels. In a phase I drug interaction study evaluating the combination of sirolimus and sorafenib in 34 patients with refractory solid tumors, best response was stable disease in 9 patients.(Gangadhar, Cohen et al. 2011) The treatment regimen was sirolimus 3 mg orally once daily (N = 20) or 2 mg twice daily (N = 14) in combination with sorafenib 400 mg twice daily. The toxicity profiles were similar to those expected for each drug alone. NCI CTCAE v3.0 grade 3 or higher toxicities occurring in at least 10% of patients in either or both treatment arms were (according to highest incidence): diarrhea in 25%, hypophosphatemia in 50%, anemia in 14%, rash in 21%, pain in 10%, hand-foot syndrome in 21%; one patient died of infection while another died of arrhythmia. There was no clinically-significant difference in Cmax for each drug individually or in combination. A phase I study of the combination of everolimus plus sorafenib in 20 patients with metastatic clear cell RCC identified maximum tolerated dosages of everolimus 5 mg daily plus sorafenib 200 mg twice daily.(Harzstark, Small et al. 2011). Five patients (25%) achieved a partial response. Grade 2 rash was observed in 55% and grade 3 rash in 10%, grade 2 or 3 hypophosphatemia in 65%, grade 2 or 3 diarrhea in 35%, grade 2 hypothyroidism in 20%, and grade 2 hypertension and hand-foot syndrome in 30% each. There was no evidence of interaction between everolimus and sorafenib on pharmacokinetic analyses in this study.

There are limited clinical data of the combination of sorafenib plus mTOR inhibition in HCC or patients with liver dysfunction. The metabolism of both sorafenib and temsirolimus is predominantly hepatic by the cytochrome P450 (CYP) 3A4 system, raising concern for drug interactions and increased toxicity with this combination, particularly in patients with underlying hepatic dysfunction. In a case report of a patient with recurrent HCC post liver transplantation who had demonstrated progression on sorafenib, the transition of immunosuppression to sirolimus and continuation of sorafenib resulted in partial response.(Wang, Speeg et al. 2010) In a series of 31 patients with HCC recurrence post liver transplantation, the transition of immunosuppression to include mTOR inhibition (everolimus or sirolimus) along with initiation of sorafenib was associated with acceptable toxicity, 1/26 (3.8%) patients with partial response, and stable disease as best response in 50%, with median overall survival of 19.3 months in this highly selected group of patients.(Gomez-Martin, Bustamante et al. 2012) In this study, everolimus, sirolimus, and sorafenib were dosed according to standard practice and investigator discretion. The most common (occurring in at least 2 patients) grade ≥ 3 toxicities were asthenia (16.1%), diarrhea (12.9%), hypertension (9.7%), gastrointestinal bleeding (6.5%), and hyperglycemia (6.5%). A phase I study of the combination of everolimus plus sorafenib in 30

patients with advanced HCC and Child-Pugh class A liver disease was presented by Finn et al at the American Society of Clinical Oncology Annual Meeting in 2011.(Finn 2011) In this study, sorafenib was dosed at 400 mg orally twice daily, with dose escalation of everolimus starting at 2.5 mg daily. Seventy-three percent of patients were Asian, and 63% had chronic active hepatitis B virus infection. The maximum tolerated dose (MTD) of everolimus in this combination was 2.5 mg daily. Dose-limiting toxicity in the 2.5 mg cohort was grade 3 transaminase elevation. In the 5 mg everolimus cohort, grade 3 or 4 thrombocytopenia occurred in 5 patients, and 1 patient had grade 3 or 4 hyperbilirubinemia. Everolimus pharmacokinetics were reported to be dose proportional and comparable to monotherapy in non-HCC patients. Best response was stable disease in 10 (62.5%) and 5 (35.7%) of patients and median time to progression was 3.5 and 3.6 months in the 2.5 and 5 mg everolimus cohorts, respectively.

1.2.3 Temsirolimus

Temsirolimus is a soluble ester analogue of sirolimus which specifically inhibits mTOR protein kinase with resulting inhibition of downstream mTOR-dependent protein translation induced by growth factor stimulation.(Dudkin, Dilling et al. 2001; Pfizer 2011) The antiproliferative effects of temsirolimus are similar to other rapalogues.(Dancey 2006) In human HCC cells, both rapamycin and temsirolimus induce growth arrest in G1 and decreased proliferation, along with reduction in phosphorylation of mTOR and p70S6K in cellular lines, and growth inhibition was in HCC PLC/PRF/5 cell xenografts in nude mice.(Hui, Tung et al. 2010) In a study of 5 cell lines including Huh7 and HepB3, temsirolimus treatment suppressed HCC cell proliferation in a dose-dependent manner, with synergistic inhibition achieved by addition of the microtubule inhibitor, vinblastine.(Zhou, Lui et al. 2012)

The metabolism of temsirolimus is predominantly hepatic via O-demethylation, hydroxylation, macrocyclic-ring opening, and ester hydrolysis to its principal metabolite, sirolimus. (Pfizer 2011) Temsirolimus is moderately bound (85% to 87%) to plasma proteins and has a large volume of distribution. There is extensive metabolism via esterase-mediated hydrolysis to the principal active metabolite, sirolimus, and to their isomers and seco-temsirolimus (M4). There are significant species-related differences in sirolimus:temsirolimus ratios thought to be due to differences in hydrolytic activity and erythrocyte concentration of FKBP-12; in humans, the sirolimus:temsirolimus ratio is 12.3. The terminal half-lives of temsirolimus and sirolimus were 17.3 hours and 54.6 hours, respectively. There was little accumulation on multiple dosing. Excretion is principally through the feces with only minor amounts (<10%) excreted in the urine.

Clearance of the mTOR inhibitor everolimus in patients with hepatic dysfunction is markedly reduced, leading to the recommendation that everolimus dosage should be reduced in the setting of impaired hepatic function.(Dancey 2006) A pharmacokinetic and pharmacodynamic study of temsirolimus in patients with advanced malignancies and varying degrees of liver dysfunction showed no significant difference in temsirolimus exposure in patients with mild to moderate liver dysfunction; in patients with Child-Pugh score of C, however, exposure to temsirolimus and its metabolite was increased.(Pfizer 2011) This study was conducted by the National Cancer Institute (NCI) Organ Dysfunction Working Group (ODWG) and was presented at the American Society of Clinical Oncology Annual Meeting in 2011.(Sarantopoulos 2011) The authors demonstrated decreased clearance, prolonged half-life, and increased AUC in patients with greater degrees of hepatic dysfunction. According to Prescribing Information, a reduction in temsirolimus starting dose to 15 mg weekly is recommended in patients with mild to moderate liver dysfunction as defined by bilirubin > 1 to 1.5 times ULN and/or AST > ULN but bilirubin within normal limits, and temsirolimus is contraindicated in patients with total bilirubin greater than 1.5 times the upper limit of normal.(Wyeth 2011)

Strong inducers of CYP3A4/5 may decrease exposure to temsirolimus and sirolimus and should be avoided. Strong inhibitors of CYP3A4 may increase exposures and should be avoided.

Moderate CYP3A4 inhibitors should be used with caution or substituted with an alternate agent. Other agents and classes with potential for drug interactions with temsirolimus are reviewed in *Section 6.7* and *Appendices 6-9 Prohibited Medications*.

Temsirolimus has been approved by the FDA for treatment of advanced renal cell carcinoma (RCC) and demonstrated a survival benefit as monotherapy by comparison with interferon alpha in a multicenter phase III trial.(Hudes, Carducci et al. 2007; Pfizer 2011) In that trial, 626 patients with previously untreated, poor prognosis, metastatic RCC were randomized to receive temsirolimus 25 mg intravenously weekly, 3 million units of interferon alpha subcutaneously three times weekly, or combination therapy with 15 mg of temsirolimus weekly plus 6 million units of interferon alpha three times weekly.(Hudes, Carducci et al. 2007) The primary endpoint, overall survival, was 10.9 months in the temsirolimus group, by comparison with 7.3 months with interferon alpha and 8.4 months with combination therapy; both overall survival and progression free survival (PFS) were significantly prolonged in the temsirolimus group by comparison with interferon alpha alone ($P = 0.008$ and $P < 0.001$, respectively).

In a phase III trial of temsirolimus in comparison with interferon alpha or combination therapy in advanced RCC, Grade 3 or 4 adverse events occurred in 68.8% of patients enrolled to the temsirolimus arm treated with 25 mg intravenously weekly.(Hudes, Carducci et al. 2007; Pfizer 2011) Adverse events (with incidence at least 5%) which were significantly more common in the temsirolimus arm versus the interferon arm were: rash (37.0% vs 5.5%), pain (27.9% vs 15.0%), hyperlipemia (27.4% vs 11.0%), hypercholesterolemia (24.5% vs 4.5%), stomatitis (19.7% vs 3.5%), pharyngitis (12.0% vs 1.5%), acne (10.1% vs 1.0%), hypokalemia (9.6% vs 3.5%), edema (9.1% vs 4.0%), rhinitis (9.6% vs 2.0%), allergic reactions (8.7% vs 0.5%), infection (6.7% vs 0.5%), face edema (6.7% vs 0.5%), and pruritic rash (5.3% vs 0.5%).(Pfizer 2011) Safety data for temsirolimus are reviewed in greater detail below in *Section 1.2.3* as well as in the Investigator's Brochure.(Pfizer 2011)

Additional information on temsirolimus is provided in *Section 4 Study Drugs* as well as in the Investigator's Brochure.(Pfizer 2011)

1.2.3.1 Phase 1 Trial of Temsirolimus plus Sorafenib in HCC

Investigators at the UCSF Helen Diller Family Comprehensive Cancer Center in partnership with the Robert H. Lurie Comprehensive Cancer Center of Northwestern University completed enrollment to a phase I clinical trial of the combination of temsirolimus and sorafenib in 25 first-line therapy for patients with advanced hepatocellular carcinoma from December 2009 and April 2012.(Kelley 2012) This study was funded by a competitive grant from the National Comprehensive Cancer Network (NCCN). Four patients remain on study as of April 2012. The MTD and recommended phase II dose (RP2D) of the combination was temsirolimus 10 mg IV weekly plus sorafenib 200 mg PO BID. The most common treatment-related grade 3 or higher toxicities by NCI CTCAE v3.0 were: Hypophosphatemia in 52%, thrombocytopenia in 24%, transaminitis in 19%, and diarrhea, fatigue, and hand-foot syndrome in 10% each. Possibly-related serious adverse events included grade 4 tumor rupture, grade 4 urosepsis, grade 3 dental infection with grade 2 neutropenia, grade 3 cellulitis with normal neutrophil count, and grade 2 pneumonia (1 event each). Confirmed partial responses were documented in 2 of 21 (10%) patients, with best response of stable disease in 11 of 21 (52%). One patient has remained on study with confirmed partial response for over 22 months as of April 2012. Among the 16 of 21 (76%) with elevated alpha-fetoprotein (AFP) tumor marker ≥ 20 ng/mL at baseline, 8 of 16 (50%) had ≥ 50% decline at the time of best AFP response.

1.2.3.2 Pharmacokinetics of Temsirolimus plus Sorafenib in Combination

In the phase I study of temsirolimus and sorafenib discussed above, preliminary results suggested that sorafenib may increase levels of the temsirolimus metabolite, rapamycin

(sirolimus), at steady state, though there were no other drug-drug interactions observed.(Patnaik A 2007) Sorafenib exposure was similar at dosages of both 200 mg and 400 mg twice daily in this study. The phase I drug interaction study evaluating the combination of sirolimus and sorafenib discussed above showed no significant interactions observed.(Gangadhar, Cohen et al. 2011) Likewise, the available data suggests no clinically relevant, measurable pharmacokinetic interaction between everolimus and sorafenib in two phase I studies of this combination in RCC (both drugs) and HCC (everolimus only).(Finn 2011; Harzstark, Small et al. 2011) Pharmacokinetic analyses for temsirolimus and its primary metabolite, sirolimus, from the UCSF-Northwestern phase I trial of temsirolimus plus sorafenib in HCC showed peak drug concentration (Cmax) for temsirolimus was similar before and after addition of sorafenib, while that of sirolimus was nonsignificantly increased. (Kelley 2012) Clearance of temsirolimus was more rapid after addition of sorafenib (5.86 L/h vs 4.37 L/h, statistical t-test, $p < 0.05$), with a nonsignificant decrease in temsirolimus half-life. The AUC inf for temsirolimus was significantly lower (statistical t-test, $p < 0.05$) after addition of sorafenib, while there was a nonsignificant trend towards increase in Cmax and AUC inf for sirolimus. There was significant interpatient variability for sirolimus across timepoints. Compared to healthy volunteers and cancer patients without liver dysfunction who were treated with 25 mg dose of temsirolimus, the Cmax for both temsirolimus and sirolimus was lower in this study which utilized a 10 mg dose.(Sarantopoulos 2011) The AUC of single-agent temsirolimus prior to addition of sorafenib in this cohort appeared similar to the non-HCC subjects treated at the 25 mg dose, and clearance was slower.

1.3 Rationale for the Proposed Study

Despite its prominence as the third most common cause of cancer death world-wide, HCC lags far behind all other malignancies with only one systemic therapy (sorafenib) with proven benefit in advanced disease. New treatments and combinations are urgently needed in this grim disease. Based upon the emerging evidence for anti-tumor activity of mTOR inhibitors in HCC, the molecular rationale for combination of mTOR inhibition with sorafenib, and the provocative data from the UCSF-Northwestern phase I trial as reported above, a phase II study is warranted to better define the efficacy of this regimen. This protocol describes a single-arm phase II study with overall goal to confirm the signal of efficacy observed in the phase I dose-finding setting, to establish the tolerability of the regimen in a larger cohort, and to further explore potential biomarkers of response. The single-arm, one-stage design will expeditiously determine whether there is an adequate signal of efficacy and adequate tolerability to warrant a much larger and significantly more costly randomized study design with single-agent sorafenib as a control arm, noting the recent negative outcomes of multiple large randomized trials in HCC (including the second-line BRISK-PS trial of brivanib versus placebo as well as the first-line trial of sunitinib versus sorafenib), whose negative results have been attributed in part to insufficient phase II data, intolerability, and lack of robust markers of response.(BusinessWire 2011; Cheng 2011)

The hypothesis of this single-arm phase II study is that the combination of temsirolimus and sorafenib will achieve a clinically-meaningful median time to progression (TTP) of at least 6 months, with null hypothesis of less than or equal to 3 months, in first-line systemic therapy for patients with advanced HCC. A randomized trial would be required to formally compare the efficacy of this combination to sorafenib alone and will be indicated if this phase II study achieves a median TTP of at least 6 months. An interim safety analysis will employ stopping rules after 30% of planned patients have been treated with at least one dose of protocol therapy to ensure the combination does not confer excessive toxicity.

A key aspect of this study will be the requirement of histologic confirmation along with adequate archival tissue for correlative tissue analyses to explore new biomarkers of response to mTOR inhibition. Circulating biomarker data including enumeration of circulating tumor cells (CTC) and measurement of the tumor marker AFP will be performed at specific timepoints to evaluate for

predictive value. Specimen banking of tissue, serum, and peripheral blood mononuclear cells will be undertaken to enable future novel biomarker studies.

1.4 Correlative Studies

This trial will include correlative studies to evaluate for potential surrogate markers of prognosis and response to therapy in HCC. In the advanced setting, such markers promise to spare patients the toxicity, opportunity loss, and financial expense of futile therapy. As new treatments become available, predictive markers may also guide changes in therapy, as well as instruct clinical trial design and facilitate the identification of new, efficacious therapies. Identification of predictive and prognostic surrogate markers may someday determine which patients will benefit from a course of adjuvant therapy after definitive resection of an early stage hepatoma, or those patients with HCC who would benefit from a strategy of maintenance therapy while awaiting transplant.

A practical limitation in prognostic and predictive assessments in HCC, however, is the paucity of tissue specimens in patients with advanced disease. Updated guidelines of the NCCN do not require biopsy in patients with classic radiographic enhancement (arterial hyperenhancement, venous washout.(NCCN 2012) These recommendations are in concordance with the American Association for the Study of Liver Diseases (AASLD) practice guidelines for radiographic diagnosis of hepatocellular carcinoma and have been validated retrospectively.(Compagnon, Grandadam et al. 2008; Bruix and Sherman 2011) Percutaneous liver biopsies often yield limited quantities of material for testing, introduce risk for significant bleeding in the setting of cirrhosis and liver dysfunction, and are at high risk for sampling error, particularly in patients with smaller tumors or with history of prior liver-directed therapies such as chemoembolization which result in necrosis. Furthermore, as with most malignancies, serial tumor tissue sampling to assess for pharmacodynamic effect or predictive factors of treatment response is not feasible. For all of these reasons, studies of tissue-based prognostication in HCC have been largely limited to hepatoma resection or explant specimens, which may differ biologically from advanced HCC. Non-invasive surrogate markers, therefore, are of particular importance in HCC.

1.4.1 Measurement of AFP as HCC Tumor Marker

Peripheral blood measurements also offer the noninvasive potential to test surrogate markers. The most commonly measured blood tumor marker of HCC is alpha-fetoprotein (AFP), a glycoprotein highly expressed during hepatocyte development.(Pang, Joh et al. 2008) AFP is secreted by approximately 70% of HCC and is diagnostic as well as prognostic.(Bruix and Sherman 2005; Pang, Joh et al. 2008; 2012) Elevated baseline AFP levels have been shown to correlate with worse prognosis after hepatic resection as well as after local therapies such as chemoembolization.(O'Suilleabhain, Poon et al. 2003; Yeh, Lee et al. 2003; Masuda T 2007) AFP level may also have predictive value; a recent study of patients enrolled in a phase III randomized trial comparing two palliative cytotoxic chemotherapy regimens showed that patients with AFP decline by at least 20% (designated as AFP responders) demonstrated markedly improved survival by comparison with AFP non-responders, with overall survival 13.5 versus 5.6 months, respectively ($p < 0.0001$).(Chan, Mo et al. 2009) AFP changes are also associated with response to sorafenib and may be an early surrogate marker of response.(Kuzuya, Asahina et al. 2011; Yau, Yao et al. 2011; Personeni, Bozzarelli et al. 2012) AFP response data were not presented in SHARP; it is notable, however, that there appeared to be a higher median baseline AFP in the placebo patients.(Llovet, Ricci et al. 2008) With the establishment of sorafenib as an effective treatment in HCC, change in AFP levels on treatment should be studied as a possible predictive marker of response in HCC.

1.4.2 Enumeration of Circulating Tumor Cells in HCC

Circulating tumor cells (CTC) will also be enumerated in patients in this study. In the majority of carcinomas, tumor cells have been detected in peripheral blood from patients by using the CellSearch System (Veridex LLC).(Allard, Matera et al. 2004) In breast and colon cancer, CTC levels convey prognostic information, correlating with PFS and overall survival; changes in CTC levels on treatment may also predict response to chemotherapy.(Cristofanilli, Budd et al. 2004; Cohen, Punt et al. 2008; Dawood, Broglio et al. 2008) In a study of 20 patients with locally advanced or metastatic hepatocellular carcinoma, CTC were detected using the CellSearch system in 9 patients (45%).(Zee BC 2007) Preliminary results from a pilot study at UCSF of 20 patients with metastatic HCC show approximately 30% to have ≥ 3 CTC/7.5mL and 47% to have ≥ 1 CTC/7.5 mL detected, while none of 10 control patients with non-malignant liver disease had detectable CTC (R. Kelley, unpublished results). A novel slide-based circulating epithelial cell assay (CEpiC) has also shown the ability to detect circulating tumor cells in patients with epithelial tumors including HCC (personal communication, P. Kuhn, The Scripps Research Institute).(Cho, Wendel et al. 2012; Wendel, Bazhenova et al. 2012) Among 5 patients treated in the PK expansion cohort of the UCSF-Northwestern phase I trial of temsirolimus plus sorafenib in advanced HCC, all 5 had detectable circulating epithelial cells by the slide-based CEpiC assay (K. Kelley, unpublished data). Portal vein tumor invasion is a negative prognostic factor in HCC, and multiple studies have shown that tumor invasion or tumor thrombosis of the portal vein is a reliable predictor of shorter survival.(Tanaka, Tobe et al. 1989; Giannelli, Pierri et al. 2002) This suggests that tumor cells within the vasculature (such as are measured by CTC testing) may also be prognostic. Along with the preliminary data showing detectability of CTC in HCC patients treated on the phase I trial of this treatment regimen, these data provide the rationale to further explore CTC levels as a potential biomarker in the current phase II study.

1.4.3 Markers of mTOR Pathway Activation

Predictive biomarkers are essential to understand the efficacy of mTOR inhibition in HCC. Activation of the mTOR pathway (e.g. underexpression or loss of PTEN and/or LKB1, and/or overexpression of pAKT and/or pS6RP) is detectable by tumor immunohistochemistry (IHC) in approximately 50% of human HCCs and appears to be related to *in vitro* sensitivity to TORC1 inhibition in HCC cell lines.[1-3] Across tumor types, there are no clinically-validated markers of response to mTOR inhibitors.[4] In a phase I/II study of everolimus in 28 patients with HCC, 3 of 11 patients with available baseline tumor specimens demonstrated activation of the mTOR pathway by IHC for pAkt (S473), pmTOR, and/or pS6RP (S235/236); the only patient with a PR in the study had moderate or high levels of each marker.[5] A prolonged partial response to everolimus in an HCC patient with high tumor pS6RP expression has been described.[6] LKB1 is a tumor suppressor which when deficient or deleted is associated with mTOR pathway activation, a metastatic phenotype and poor outcomes.[7] Preclinical models of LKB1-deficient tumors suggest sensitivity to mTOR inhibition.[7] A correlative objective will be to examine whether tumor mTOR pathway activation as measured by selected IHC markers and by tumor next-generation DNA sequencing of a targeted panel including mTOR pathway genes is related to clinical outcomes of TORC1 inhibition.

1.4.4 Reactivation of Viral Hepatitis B Infection

Given the potential immunosuppressive effect of mTOR inhibition, monitoring for viral hepatitis reactivation or exacerbation will be undertaken in patients with chronic active HBV or HBV carriers undergoing experimental therapy with temsirolimus plus sorafenib.

1.4.5 Cause of Underlying Liver disease

As discussed in the previous sections, disparate outcomes are obtained with the use of sorafenib for HCC in a Western patient population with greater proportion having underlying HCV infection, by comparison to an Asian patient population with greater proportion having underlying HBV infection.(Cheng A. 2008; Kelley and Venook 2008; Llovet, Ricci et al. 2008) For this reason, subgroup analyses to compare TTP, PFS, OS, response rate, AFP response, and toxicity and tolerability based upon cause of underlying liver disease as well as Asian versus non-Asian ethnicity will be performed as exploratory endpoints.

1.4.6 Specimen Banking

Studies in archival hepatoma tissue have suggested the prognostic value of a host of molecular features of HCC. These include overexpression of angiogenic factors, activation of the Ras/Raf/ERK/MEK pathway, and aberrant mTOR pathway signaling.(Kim RD 2008; Li, Tan et al. 2008; Schmitz, Wohlschlaeger et al. 2008; Villanueva, Chiang et al. 2008; Baba, Wohlschlaeger et al. 2009) In resected hepatoma tissue specimens, activation of components of the mTOR pathway including AKT and p70S6K correlate with a more invasive phenotype and poorer prognosis.(Li, Tan et al. 2008; Schmitz, Wohlschlaeger et al. 2008; Villanueva, Chiang et al. 2008) Down-regulation of the tumor-suppressor PTEN which regulates mTOR activation may also confer poorer prognosis.(Villanueva, Chiang et al. 2008) The success of sorafenib in HCC raises the question of whether tissue markers of Ras/Raf/ERK/MEK pathway activation may also predict response to this agent. In the phase II study of sorafenib in HCC, higher pretreatment tumor phosphorylated ERK (pERK) levels within the nucleus, as measured by immunohistochemistry (IHC) in 33 patients, appeared to correlate with longer time to progression, though this association was not observed among the 110 patients with tissue pERK IHC testing from the SHARP trial.(Abou-Alfa, Schwartz et al. 2006; Llovet 2012) In another study of liver resection or explant specimens, increased levels of cytoplasmic pERK by IHC appeared to correlate with poorer prognosis and were associated with HCV infection, though this study did not involve patients treated with sorafenib.(Schmitz, Wohlschlaeger et al. 2008) Preclinical studies suggest that loss of PTEN activity and increased levels of activated AKT may confer sensitivity to mTOR inhibitors in vitro.(Neshat, Mellinghoff et al. 2001; Podsypanina, Lee et al. 2001) The degree and duration of p70S6K inhibition in PBMCs may also correlate with response to mTOR inhibitors.(Peralba, DeGraffenried et al. 2003; Boulay, Zumstein-Mecker et al. 2004)

Although we do not expect to have adequate numbers or quantities of tissue specimens to perform molecular testing in the context of this trial, we do plan to bank optional patient blood specimens (including serum, PBMC, whole blood, and plasma) specimens as well as fixed left-over tissue specimens when available from all enrolled patients in this trial for possible future molecular, pharmacogenomic, and/or proteomic testing. Such testing would be indicated if there is a subset of patients with sustained radiographic response and/or prolonged disease control to suggest the existence of underlying predictive biomarkers.

2. Objectives of the Study

2.1 Primary

- To determine the median time to progression (TTP) in patients with advanced HCC treated with combination of temsirolimus plus sorafenib

2.2 Secondary

- To determine response rate (RR) using RECIST 1.1
- To measure median progression-free survival (PFS)

- To measure median overall survival (OS)
- To measure time to treatment failure (TTF)
- To measure change in AFP tumor marker and examine for association between change in AFP and TTP and best response
- To characterize toxicity and tolerability of the combination of temsirolimus plus sorafenib, including describing frequency of dose reductions, delays, and discontinuation for toxicity

2.3 Exploratory Objectives and Other Assessments

- TTP, RR, PFS, OS, TTF, AFP response, and toxicity and tolerability will be described in subsets defined by HBV, HCV, Asian, and non-Asian status.
- To enumerate CTC at baseline and after 1 and 2 cycles of therapy to evaluate for any relationship between baseline levels and change in levels with median TTP and/or OS
- To characterize baseline mTOR pathway activation by immunohistochemistry (IHC) for selected markers such as pS6RP, pAKT, PTEN, and LKB1, and by tumor next-generation DNA sequencing of targeted genes including mTOR pathway in archival tumor specimens from HCC patients treated with temsirolimus plus sorafenib.
- To examine the relationship between baseline mTOR pathway activation by IHC and by sequencing and clinical outcomes on treatment with temsirolimus plus sorafenib.
- In patients with chronic active HBV infection or exposure, to monitor hepatitis viral load during and after therapy with an mTOR inhibitor
- To bank peripheral blood specimens and left-over archival pathology or cytology specimens for future research

2.4 Endpoints

2.4.1 Primary Endpoint

- Median TTP will be calculated in months from date of first dose of protocol therapy to date of removal from study for progression. For patients removed from study for other reasons than progression (such as toxicity, withdrawal of consent, death without documented progression, or other reasons without clinical or radiographic evidence of tumor progression), TTP will be censored at the date last known to be progression-free.

2.4.2 Secondary Endpoints

- Response rate (RR) will be measured by RECIST version 1.1.
- Median PFS will be calculated in months from date of first dose of protocol therapy to date of documented disease progression or death from any cause. For patients removed from study for other reasons than progression or death, PFS will be censored at the date last known to be progression-free.
- Median OS for all enrolled patients who receive at least one dose of protocol therapy will be calculated from date of first dose of protocol therapy until date of death, using chart review and/or follow up phone calls to determine date of death in patients after removal from study.
- TTF will be measured from date of first dose of protocol therapy to date of study discontinuation for progression, death, or adverse events (related or unrelated). For patients removed from study for other reasons (such as noncompliance or withdrawal of consent), TTF will be censored at the date of study discontinuation.
- In patients with baseline AFP ≥ 20 ng/mL, AFP response will be measured by the percent change from baseline value to the value at the time of best AFP response. The proportion

of ≥ 50% decline from baseline will be measured. The change in AFP will also be examined for association with absolute TTP and best response.

- Toxicity will be measured using Common Terminology Criteria for Adverse Events (CTCAE) version 4.0. The frequency of dose reduction, treatment delays, and discontinuation for lack of tolerability will be quantified.

2.4.3 Exploratory Endpoints

- TTP, RR, PFS, OS, TTF, AFP response, and toxicity and tolerability will be described in subsets defined by HBV, HCV, Asian, and non-Asian status.
- CTC will be enumerated in peripheral blood at baseline and after 1 and 2 cycles of therapy to evaluate for any relationship between baseline levels and change in levels with median TTP and/or OS.
- Baseline mTOR pathway activation status as measured by immunohistochemistry (IHC) for selected markers such as pS6RP, pAKT, PTEN, and LKB1, and by a targeted next-generation tumor DNA sequencing panel including mTOR pathway genes in archival tumor specimens.
- Relationship between baseline mTOR pathway activation by IHC and/or sequencing and clinical outcomes including TTP and AFP response.
- In patients with chronic active HBV and/or positive HBcAb, hepatitis B viral DNA quantitative will be monitored for rates of viremia/reactivation on study.
- Optional patient blood specimens (including serum, PBMC, whole blood, and plasma) and left-over archival pathology or cytology specimens will be banked in the UCSF Hepatobiliary Tissue Bank for future research.

3. Study Design

3.1 Characteristics

This study is a single-arm, one-stage, open-label phase II trial to evaluate the efficacy of the combination of temsirolimus plus sorafenib in first line therapy for patients with histologically-confirmed advanced HCC.

3.2 Number of Subjects

The target sample size is 25 evaluable patients for the primary endpoint. With the expectation of approximately 10% ineligibility rate, the expected maximum enrollment will be 28 patients. Eligible patients who receive at least one dose of protocol therapy will not be replaced. Patients will be replaced and removed from study if determined to be ineligible after enrollment (for reasons such as inability to comply/withdrawal of consent or other ineligibility)

3.3 Eligibility Criteria

The study population will be patients with advanced, unresectable HCC with adequate performance status and Child-Pugh class A or B with ≤ 7 points liver function who have not received prior systemic therapy including no prior sorafenib.

Patients must have baseline evaluations performed prior to the first dose of study drug and must meet all inclusion and exclusion criteria. In addition, the patient must be thoroughly informed about all aspects of the study, including the study visit schedule and required evaluations and all regulatory requirements for informed consent. The written informed consent must be obtained from the patient prior to enrollment. The following criteria apply to all patients enrolled onto the study unless otherwise specified.

3.3.1 Inclusion Criteria

1. Patients must have histologically diagnosed AJCC stage II, III, or IV HCC not eligible for curative resection, transplantation, or ablative therapies
    a. Cases with mixed, composite, or combined HCC-cholangiocarcinoma histology are eligible with approval from study chair and provided the treating investigator believes it is in the best interest of the patient to treat the tumor with therapy targeted towards the HCC component of tumor based upon review of pathology and clinical characteristics
2. Radiographically measurable disease by RECIST version 1.1 in at least one site not previously treated with chemoembolization, radioembolization, or other local ablative procedures (i.e. must have at least one measurable target lesion, either within the liver or in a measurable metastatic site); a new area of tumor progression within or adjacent to a previously-treated lesion, if clearly measurable by a Radiologist, is acceptable
3. No prior systemic cytotoxic chemotherapy or targeted therapy (including sorafenib) for HCC
4. Prior chemoembolization, local ablative therapies, or hepatic resection permitted if completed ≥ 4 weeks prior to study enrollment if patient has recovered with ≤ grade 1 toxicity and if measurable disease (criterion 2) is present
5. Prior radiation for bone or brain metastases is permitted if patient is now asymptomatic and has completed all radiation and steroid therapy (if applicable) for brain or bone metastases ≥ 2 weeks prior to study enrollment.
6. Age ≥ 18 years.
7. Child-Pugh score of A or B with ≤ 7 points and meeting laboratory eligibility for all parameters
8. ECOG performance status of 0 or 1
9. Life expectancy greater than 3 months
10. Treatment with appropriate antiviral therapy for patients with active HBV infection is required
11. Treatment for clinically-significant hyperglycemia, hyperlipidemia, or hypertension that develops on study is required
12. Baseline blood pressure must be adequately controlled with or without antihypertensive medications prior to enrollment (systolic ≤ 150 mm Hg, diastolic ≤ 90 mm Hg)
13. Baseline cholesterol must be < 350 mg/dL and triglycerides < 300 mg/dL (with or without the use of antihyperlipidemic medications)
14. Baseline fasting blood glucose must be ≤ 140 mg/dL and hemoglobin A1c less than 7.5% (with or without the use of anti-diabetic medications)
15. Adequate baseline organ and marrow function as defined below

    <u>Adequate bone marrow function:</u>

absolute neutrophil count	≥ 1,000/mcL
platelets	≥ 75,000/mcL

hemoglobin	≥ 8.5 g/dL
Adequate hepatic function:	
total bilirubin	≤ 2 mg/dL or ≤ 1.5 times ULN
AST(SGOT) & ALT (SGPT)	≤ 5 X ULN
INR	≤1. 5 X ULN
Adequate renal function:	
albumin	≥ 2.8 g/dL
creatinine	≤1. 5 X ULN

16. Able to tolerate oral therapy.

17. Ability to understand and willingness to provide informed consent, and the willingness to comply with the requirements of the protocol. Informed consent may be obtained with the assistance of a medical translator according to institutional policies.

18. The effects of temsirolimus on the developing human fetus are unknown. For this reason and because sorafenib - also being used in this trial - is known to be teratogenic, women of child-bearing potential must have a negative pregnancy test within 14 days of study enrollment.

    Also, women of child-bearing potential and men must agree to use two methods adequate contraception (hormonal plus barrier or two forms of barrier) or abstinence prior to study entry and for the duration of study participation. Should a woman become pregnant or suspect she is pregnant while she or her partner is participating in this study, she needs to inform her treating physician immediately. Men treated or enrolled on this protocol must also agree to use adequate contraception prior to the study, or be surgically sterile, for the duration of study participation, and for 3 months after completion of study drug administration.

19. Eligibility of patients receiving any medications or substances known to affect or with potential to affect the activity or pharmacokinetics of temsirolimus and/or sorafenib will be determined following review of the case by the Study Chair. Efforts should be made to switch patients who are taking enzyme-inducing anti-convulsant agents to other medications. A list of medications and substances known or with the potential to interact with selected relevant CYP450 isoenzymes, P-glycoprotein pathway, and/or UGT1A1 glucuronidation is provided in *Appendices 6-9*.

3.3.2 Exclusion Criteria

1. Biliary tract cancers or fibrolamellar variant tumors are excluded.

    a. Cases with mixed, composite, or combined HCC-cholangiocarcinoma histology are eligible with approval from study chair and provided the treating investigator believes it is in the best interest of the patient to treat the tumor with therapy targeted towards the HCC component of tumor based upon review of pathology and clinical characteristics

2. Prior systemic or antiangiogenic therapy for HCC (including thalidomide, sorafenib, sunitinib, or bevacizumab). Prior systemic therapy for other diagnoses is permitted if greater than 6 months have elapsed since last dose, any prior toxicity has recovered to ≤ grade 1 by CTCAE v4.0, and treatment was not discontinued for toxicity.

3. Prior treatment with mTOR inhibitor or other molecularly targeted therapy.\

4. Prior systemic cytotoxic therapies for HCC (chemoembolization is permitted if inclusion criteria are met).

5. Treatment with other investigational agents.

6. Immunosuppressive medications including systemic corticosteroids unless used for adrenal replacement, appetite stimulation, acute therapy for asthma or bronchitis exacerbation (≤ 2 weeks), or antiemesis

7. Patients with known HIV infection are ineligible due to risk of pharmacokinetic interactions between anti-retroviral therapy and the study drugs, as well as potential for significant immunosuppression and serious infections with mTOR inhibition.

8. Patients who have undergone liver transplantation are excluded.

9. Uncontrolled hypertension (> 150/90 mmHg).

10. Uncontrolled hyperlipidemia (total cholesterol > 350 or triglycerides > 300).

11. Symptomatic brain or bone metastases; prior radiation and/or steroid therapy for brain or bone metastases (if applicable) must be completed ≥ 2 weeks prior to study enrollment.

12. History of seizure disorder requiring antiepileptic medication or brain metastases with seizures.

13. Serious non-healing wound, ulcer, bone fracture, or abscess.

14. Major surgical procedure less than 4 weeks from start of protocol treatment.

15. Patients requiring chronic anticoagulation with warfarin are excluded. Patients treated with low molecular weight heparin or unfractionated heparin are eligible if on a stable dose without evidence of clinically significant bleeding for at least 2 weeks prior to enrollment.

16. Active second malignancy other than non-melanoma skin cancer or cervical carcinoma in situ. (Patients with history of malignancy are not considered to have a “currently active” malignancy if they have completed therapy and are now considered by their physician to be at less than 30% risk for relapse.)

17. Uncontrolled intercurrent illness including, but not limited to: Ongoing or active infection, symptomatic congestive heart failure, unstable angina pectoris, uncontrolled cardiac arrhythmia, uncontrolled peripheral vascular disease, myocardial infarction within preceding 12 months, cerebrovascular accident within preceding 12 months, pulmonary disease impairing functional status or requiring oxygen, impairment in gastrointestinal function that may affect or alter absorption of oral medications (such as malabsorption or history of gastrectomy or bowel resection).

18. Patients will be excluded if there is any history of allergic reaction(s) attributed to compounds of similar composition to temsirolimus, sorafenib, their metabolites, or any component of their formulation (including excipients and polysorbate 80). This includes hypersensitivity to macrolide antibiotics due to potential for cross-reactivity with temsirolimus.

19. Pregnant or lactating women are excluded from this study because temsirolimus and sorafenib are drugs with 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 temsirolimus or sorafenib, breastfeeding should be discontinued if the mother is receiving temsirolimus/sorafenib treatment.

20. Patients who require prohibited medications with potential for serious interactions with protocol therapy, and who cannot have therapeutic substitution are excluded. Patients receiving any medications or substances that are inhibitors or inducers of CYP450 enzyme(s) are ineligible. Lists of prohibited medications and substances known, or with the potential to interact with the specified CYP450 enzyme(s) isoenzymes are provided in *Appendices 6-9 Prohibited Medications*

21. Psychiatric illness, other significant medical illness, or social situation which, in the investigator's opinion, would limit compliance or ability to comply with study requirements.

3.4 Duration of Therapy

In the absence of treatment delays due to adverse events, individual patient treatment may continue indefinitely, or until:

- Disease progression by RECIST 1.1 or clinically
- Inter-current illness that prevents further administration of treatment
- Unacceptable adverse event(s) toxicity, or unmanageable toxicity despite optimal supportive care and dose modifications *(Section 5 Treatment Plan)*
- Patients decides to withdraw from the study or is unable to comply with study procedures
- General or specific changes in the patients' condition, or other factors which may influence safety or compliance, that render the patient unacceptable for further treatment in the judgment of the investigator

When a patient is discontinued from the study, the reason(s) for discontinuation will be documented in study records.

3.5 Duration of Follow Up

After treatment discontinuation, follow up for overall survival monitoring will be performed as described in *Section 6.1 Schedule of Procedures and Observations* for up to 5 years or until death, whichever comes first. The study will end 5 years after the final patient discontinues treatment or after all patients are deceased, whichever comes first.

3.6 Randomization Procedures

This study does not involve randomization

3.7 Study Timeline

3.7.1 Primary Completion

Based upon the accrual rate to the predecessor phase I trial of this regimen at UCSF and Northwestern, the study is expected to complete accrual approximately 24 months from the time the study opens to accrual.

3.7.2 Study Completion

The study is expected to reach completion once the final patient has been enrolled and all patients have had potential for at least 6 months of follow up or been removed from study. Study completion is expected approximately 2.5 years from study opening.

4. Study Drugs

4.1 Description, Supply and Storage of Investigational Drugs

4.1.1 Temsirolimus (TORISEL®)

Please refer to the updated Investigator's Brochure as well as Package Insert for temsirolimus for complete drug information. (Pfizer 2011; Wyeth 2011)

Temsirolimus is supplied as a kit containing TORISEL® injection vial and a diluents vial. The TORISEL® vial contains temsirolimus at a concentration of 25 mg/mL. The vial contains an overfill of 0.2 mL to ensure the ability to withdraw the recommended dose. The diluent vial includes a deliverable volume of 1.8 mL. This vial contains an overfill in order to ensure that the appropriate volume can be withdrawn.

Classification

Temsirolimus is a specific inhibitor of the mammalian target of rapamycin (mTOR), an enzyme that regulates cell growth and proliferation.

Mechanism of Action

Temsirolimus is an inhibitor of mTOR (mammalian target of rapamycin). Temsirolimus binds to an intracellular protein (FKBP-12), and the protein-drug complex inhibits the activity of mTOR that controls cell division. Inhibition of mTOR activity resulted in a G1 growth arrest in treated tumor cells. When mTOR was inhibited, its ability to phosphorylate p70S6k and S6 ribosomal protein, which are downstream of mTOR in the PI3 kinase/AKT pathway was blocked. In *in vitro* studies using renal cell carcinoma cell lines, temsirolimus inhibited the activity of mTOR and resulted in reduced levels of the hypoxia-inducible factors HIF-1 and HIF-2 alpha, and the vascular endothelial growth factor (VEGF).

Metabolism

Cytochrome P450 3A4 is the major isozyme responsible for the formation of five temsirolimus metabolites. Sirolimus, an active metabolite of temsirolimus, is the principal metabolite in humans following intravenous treatment. The remainder of the metabolites account for less than 10% of radioactivity in the plasma. In human liver microsomes temsirolimus was an inhibitor of CYP2D6 and 3A4. However, there was no effect observed in vivo when temsirolimus was administered with desipramine (a CYP2D6 substrate), and no effect is anticipated with substrates of CYP3A4 metabolism.

Elimination is primarily via the feces. After a single IV dose of [14C]-temsirolimus approximately 82% of total radioactivity was eliminated within 14 days, with 4.6% and 78% of the administered radioactivity recovered in the urine and feces, respectively. Following a single 25 mg dose of TORISEL® in patients with cancer, temsirolimus mean (CV) systemic clearance was 16.2 (22%) L/h. Temsirolimus exhibits a bi-exponential decline in whole blood concentrations and the mean half-lives of temsirolimus and sirolimus were 17.3 hr and 54.6 hr.

Temsirolimus is a substrate of the efflux transporter P-glycoprotein (Pgp) in vitro. If TORISEL® is administered with drugs that inhibit Pgp, increased concentrations of temsirolimus are likely, and caution should be exercised. *In vitro*, temsirolimus inhibited human Pgp (IC50 value of 2μM). If TORISEL® is administered with drugs that are substrates of Pgp, increased concentrations of the substrate drug are likely and caution should be exercised.

No dosage adjustment for temsirolimus is recommended in patients with renal impairment. There have been no clinical studies of this agent in patients with impaired renal function, though renal impairment is not expected to markedly influence drug exposure. Less than 5% of total radioactivity was excreted in the urine following a 25 mg intravenous dose of [14C]-labeled

temsirolimus in healthy subjects. Temsirolimus has not been studied in patients undergoing hemodialysis.

Temsirolimus is cleared predominantly by the liver.

Contraindications

Temsirolimus IV is contraindicated in persons with known hypersensitivity to temsirolimus or any component of the formulation. Temsirolimus is contraindicated in patients with bilirubin >1.5 x ULN.

Availability

Temsirolimus is FDA-approved for the treatment of advanced renal cell carcinoma (RCC). Temsirolimus will be supplied for this trial directly by the manufacturer, Pfizer, Inc., from commercial supplies. See *Section 4.2 Drug Accountability*.

Storage and handling

Temsirolimus Concentrate for Injection should be stored refrigerated (2° C to 8° C) and protected from light. When co-packaged with Temsirolimus Concentrate for Injection, Diluent for Temsirolimus Concentrate for Injection should be stored refrigerated (2° C to 8° C). The active product and diluent must be allowed to warm to room temperature for approximately 1 hour before dilution. Dilution of Temsirolimus Concentrate for Injection in Diluent for Temsirolimus Concentrate for Injection must be followed by further dilution into an infusion bag or bottle of 0.9% sodium chloride injection. The drug-diluent mixture is stable for up to 24 hours at controlled room temperature.

The final diluted infusion solution (drug-diluent in sodium chloride injection) should be stored in a secured, clean environment, at room temperature, and administered within 6 hours from the time that the concentrate-diluent mixture is added to the 0.9% sodium chloride injection.

Admixtures of temsirolimus are stable under ordinary fluorescent room light, but should be protected from excessive light, such as sunlight. Temsirolimus Concentrate for Injection, when constituted, contains polysorbate 80, which is

known to increase the rate of di-(2-ethylhexyl)phthalate (DEHP) extraction from polyvinyl chloride (PVC). This should be considered during the preparation and administration of Temsirolimus Concentrate for Injection admixture, including storage time elapsed when in direct contact with PVC following constitution. It is recommended to use infusion bags and sets composed of glass, polyolefin, or polyethylene, to avoid excessive loss of drug and to decrease the rate of di-(2-ethylhexyl)phthalate (DEHP) extraction.

An in-line filter with a pore size of not greater than 5 microns is recommended for administration. The following are examples of in-line filters that are compatible with Temsirolimus Concentrate for Injection:

- IV 6200 Disposable IV Filter 0.2 μm, by EPS, Inc.
- IV 6120 Disposable IV Filter 1.2 μm, by EPS, Inc.
- LV 5000 Large Volume 5 μm Conical Filter, by B. Braun
- Codan 0.2 μm monofilter
- Baxter Paclitaxel Set with 0.22 μm filter

Other filters may be used if the filter medium is composed of polyethersulfone

Refer to the temsirolimus Package Insert for updated and complete storage and handling information.

Preparation

During handling and preparation of admixtures, temsirolimus should be protected from excessive room light and sunlight. Parenteral drug products should be inspected visually for particulate matter and discoloration prior to administration, whenever solution and container permit. In order to minimize the patient exposure to the plasticizer DEHP (di-2-ethylhexyl phthalate), which may be leached from PVC infusion bags or sets, the final temsirolimus dilution for infusion should be stored in bottles (glass, polypropylene) or plastic bags (polypropylene, polyolefin) and administered through polyethylene-lined administration sets.

Dilution: In preparing the TORISEL® administration solution, follow this two-step dilution process in an aseptic manner.

Step 1: Inject 1.8 mL of DILUENT for TORISEL® into the vial of TORISEL® (temsirolimus) injection (25 mg/ml). The TORISEL® (temsirolimus) vial contains an overfill of 0.2 mL (30 mg/1.2 mL). Due to the intentional overfill in the TORISEL® injection vial, the drug concentration of the resulting solution will be 10 mg/mL. A total volume of 3 mL will be obtained including the overfill. Mix well by inversion of the vial. Allow sufficient time for air bubbles to subside. This 10 mg/mL drug solution/diluent mixture must be further diluted as described in Step 2 below. The solution is clear to slightly turbid, colorless to yellow, and free from visual particulates. The 10 mg/mL drug solution/diluent mixture is stable for up to 24 hours at controlled room temperature.

Step 2: Withdraw the required amount of temsirolimus from the 10 mg/mL drug solution/diluent mixture prepared in Step 1. Inject rapidly into a 250 mL container (glass, polyolefin, or polyethylene) of 0.9% sodium chloride injection. Mix the admixture by inversion of the bag or bottle. Avoid excessive shaking as this may cause foaming.

Administration

Administration of the final diluted infusion solution should be completed within six hours from the time that the concentrate diluent mixture is added to the sodium chloride injection.

Temsirolimus Concentrate for Injection is infused over a 30-60 minute period (60 minutes for all doses during cycle one) once a week. The use of an infusion pump is the preferred method of administration to ensure accurate delivery of the drug. It is recommended to use infusion bags and sets composed of glass, polyolefin, or polyethylene, to avoid excessive loss of drug and to decrease the rate of di-(2-ethylhexyl) phthalate (DEHP) extraction. This should be considered during the preparation and administration of Temsirolimus Concentrate for Injection admixture, including storage time elapsed when in direct contact with PVC following constitution. An in-line filter with a pore size of not greater than 5 microns is recommended for administration.

Incompatibilities

Undiluted TORISEL® injection should not be added directly to aqueous infusion solutions. Direct addition of TORISEL® injection to aqueous solutions will result in precipitation of drug. Always combine TORISEL® injection with DILUENT for TORISEL® before adding to infusion solutions. It is recommended that TORISEL® be administered in 0.9% sodium chloride injection after combining with diluent. The stability of TORISEL® in other infusion solutions has not been evaluated. Addition of other drugs or nutritional agents to admixtures of TORISEL® in sodium chloride injection has not been evaluated and should be avoided. Temsirolimus is degraded by both acids and bases, and thus combinations of temsirolimus with agents capable of modifying solution pH should be avoided.

Side Effects

Complete and updated adverse event information is available in the Investigator's Brochure and/or Package Insert for temsirolimus (TORISEL®). (Pfizer 2011; Wyeth 2011)

The most common adverse reactions (incidence ≥ 30%) are rash, asthenia, mucositis, nausea, edema, and anorexia. The most common laboratory abnormalities (incidence ≥ 30%) are anemia, hyperglycemia, hyperlipemia, hypertriglyceridemia, elevated alkaline phosphatase, elevated serum creatinine, lymphopenia, hypophosphatemia, thrombocytopenia, elevated AST, and leukopenia. Specific adverse events which may be associated with temsirolimus and/or special precautions for its use include but are not limited to:

Hypersensitivity Reactions

Hypersensitivity/infusion reactions, including but not limited to flushing, chest pain, dyspnea, hypotension, apnea, loss of consciousness, hypersensitivity and anaphylaxis, have been associated with the administration of temsirolimus. These reactions can occur very early in the first infusion, but may also occur with subsequent infusions. Patients should be monitored throughout the infusion and appropriate supportive care should be available. Temsirolimus infusion should be interrupted in all patients with severe infusion reactions and appropriate medical therapy administered. temsirolimus should be used with caution in persons with known hypersensitivity to temsirolimus or its metabolites (including sirolimus), polysorbate 80, or to any other component (including the excipients) of temsirolimus. An H1 antihistamine should be administered to patients before the start of the intravenous temsirolimus infusion. Temsirolimus should be used with caution in patients with known hypersensitivity to an antihistamine, or patients who cannot receive an antihistamine for other medical reasons. If a patient develops a hypersensitivity reaction during the temsirolimus infusion, the infusion should be stopped and the patient should be observed for at least 30 to 60 minutes (depending on the severity of the reaction). At the discretion of the physician, treatment may be resumed with the administration of an H1-receptor antagonist (such as diphenhydramine), if not previously administered, and/or an H2-receptor antagonist (such as intravenous famotidine 20 mg or intravenous ranitidine 50 mg) approximately 30 minutes before restarting the temsirolimus infusion. The infusion may then be resumed at a slower rate (up to 60 minutes). A benefit-risk assessment should be done prior to the continuation of temsirolimus therapy in patients with severe or life-threatening reactions.

Hyperglycemia/Glucose Intolerance

The use of temsirolimus is likely to result in increases in serum glucose. In the phase 3 trial, 89% of patients receiving temsirolimus had at least one elevated serum glucose while on treatment, and 26% of patients reported hyperglycemia as an adverse event. This may result in the need for an increase in the dose of, or initiation of, insulin and/or oral hypoglycemic agent therapy. Serum glucose should be tested before and during treatment with temsirolimus. Patients should be advised to report excessive thirst or any increase in the volume or frequency of urination.

Infections

The use of temsirolimus may result in immunosuppression. Patients should be carefully observed for the occurrence of infections, including opportunistic infections.

Interstitial Lung Disease

Cases of interstitial lung disease, some resulting in death, occurred in patients who received temsirolimus. Some patients were asymptomatic, or had minimal symptoms, with infiltrates detected on computed tomography scan or chest radiograph. Others presented with symptoms such as dyspnea, cough, hypoxia, and fever. Some patients required discontinuation of temsirolimus and/or treatment with corticosteroids and/or antibiotics, while some patients continued treatment without additional intervention. Patients should be advised to report promptly any new or worsening respiratory symptoms. It is recommended that patients undergo baseline radiographic assessment by lung computed tomography scan or chest radiograph prior to the initiation of temsirolimus L therapy. Follow such assessments periodically, even in the

absence of clinical respiratory symptoms. It is recommended that patients be followed closely for occurrence of clinical respiratory symptoms. If clinically significant respiratory symptoms develop, consider withholding temsirolimus administration until after recovery of symptoms and improvement of radiographic findings related to pneumonitis. Empiric treatment with corticosteroids and/or antibiotics may be considered.

Hyperlipemia

The use of temsirolimus is likely to result in increases in serum triglycerides and cholesterol. In the phase 3 trial, 87% of patients receiving temsirolimus had at least one elevated serum cholesterol value and 83% had at least one elevated serum triglyceride value. This may require initiation, or increase in the dose, of lipid-lowering agents. Serum cholesterol and triglycerides should be tested before and during treatment with temsirolimus.

Bowel Perforation

Cases of fatal bowel perforation occurred in patients who received TORISEL®. These patients presented with fever, abdominal pain, metabolic acidosis, bloody stools, diarrhea, and/or acute abdomen. Patients should be advised to report promptly any new or worsening abdominal pain or blood in their stools.

Renal Failure

Cases of rapidly progressive and sometimes fatal acute renal failure not clearly related to disease progression occurred in patients who received temsirolimus. Some of these cases were not responsive to dialysis.

Wound Healing Complications

Use of temsirolimus has been associated with abnormal wound healing. Therefore, caution should be exercised with the use of TORISEL® in the perioperative period.

Intracerebral Hemorrhage

Patients with central nervous system tumors (primary CNS tumor or metastases) and/or receiving anticoagulation therapy may be at an increased risk of developing intracerebral bleeding (including fatal outcomes) while receiving temsirolimus L.

Thrombocytopenia and Neutropenia

Grades 3 and 4 thrombocytopenia and neutropenia have been observed in subjects with mantle cell lymphoma.

Local Injection Site Reactions

There are also postmarketing reports of temsirolimus extravasations resulting in swelling, pain, warmth, and erythema.

Stevens-Johnson Syndrome

This life-threatening skin rash has been reported in patients who received temsirolimus.

Rare but Serious Events

The following adverse reactions have been identified during post approval use of temsirolimus. Because these reactions are reported voluntarily from a population of uncertain size, it is not always possible to readily estimate their frequency or establish a causal relationship to drug exposure. The following adverse reactions have been observed in patients receiving temsirolimus: pleural effusion, hemodynamically significant pericardial effusions requiring intervention, convulsions, rhabdomyolysis, Stevens-Johnson Syndrome, and complex regional pain syndrome (reflex sympathetic dystrophy).

Co-administration with Inducers or Inhibitors of CYP3A Metabolism

Agents Inducing CYP3A Metabolism: Strong inducers of CYP3A4/5 such as dexamethasone, carbamazepine, phenytoin, phenobarbital, rifampin, rifabutin, and rifampacin may decrease exposure of the active metabolite, sirolimus. If alternative treatment cannot be administered, a dose adjustment should be considered. St. John's Wort may decrease temsirolimus plasma concentrations unpredictably. Patients receiving temsirolimus should not take St. John's Wort concomitantly.

Agents Inhibiting CYP3A Metabolism: Strong CYP3A4 inhibitors such as atazanavir, clarithromycin, indinavir, itraconazole, ketoconazole, nefazodone, nelfinavir, ritonavir, saquinavir, and telithromycin may increase blood concentrations of the active metabolite sirolimus. If alternative treatments cannot be administered, a dose adjustment should be considered.

Concomitant use of temsirolimus with sunitinib

The combination of temsirolimus and sunitinib resulted in dose-limiting toxicity. Dose-limiting toxicities (Grade 3/4 erythematous maculopapular rash, and gout/cellulitis requiring hospitalization) were observed in two out of three patients treated in the first cohort of a phase 1 study at doses of temsirolimus 15 mg IV per week and sunitinib 25 mg oral per day (Days 1-28 followed by a 2-week rest).

Administration of Vaccinations

The use of live vaccines and close contact with those who have received live vaccines should be avoided during treatment with temsirolimus. Examples of live vaccines are: intranasal influenza, measles, mumps, rubella, oral polio, BCG, yellow fever, varicella, and TY21a typhoid vaccines.

Use in Pregnancy

Pregnancy Category D. Temsirolimus administered daily as an oral formulation caused embryo-fetal and intrauterine toxicities in rats and rabbits at human sub-therapeutic exposures. Embryo-fetal adverse effects in rats consisted of reduced fetal weight and reduced ossifications, and in rabbits included reduced fetal weight, omphalocele, bifurcated sternabrae, notched ribs, and incomplete ossifications. In rats, the intrauterine and embryo-fetal adverse effects were observed at the oral dose of 2.7 mg/m2/day (approximately 0.04-fold the AUC in cancer patients at the human recommended dose). In rabbits, the intrauterine and embryo-fetal adverse effects were observed at the oral dose of ≥7.2 mg/m2/day (approximately 0.12-fold the AUC in cancer patients at the recommended human dose). Women of childbearing potential should be advised to avoid becoming pregnant throughout treatment and for 3 months after TORISEL® therapy has stopped. Temsirolimus can cause fetal harm when administered to a pregnant woman. If this drug is used during pregnancy, or if the patient becomes pregnant while taking this drug, the patient should be apprised of the potential hazard to the fetus. Men should be counseled regarding the effects of TORISEL® on the fetus and sperm prior to starting treatment. Men with partners of childbearing potential should use reliable contraception throughout treatment and are recommended to continue this for 3 months after the last dose of TORISEL®.

Use in Breastfeeding

It is not known whether temsirolimus is excreted into human milk. Animal studies suggest potential for tumorigenicity for sirolimus (active metabolite of temsirolimus) in animal studies. Breastfeeding patients are excluded from this study.

Use in Elderly Patients

Based on the results of a phase 3 study, elderly patients may be more likely to experience certain adverse reactions including diarrhea, edema and pneumonia.

4.1.2 Sorafenib (NEXAVAR®)

Please refer to the Investigator's Brochure and Package Insert for sorafenib for complete information. (Bayer 2011)

NEXAVAR® is prepared as tablets containing sorafenib tosylate (274 mg) equivalent to 200 mg of sorafenib. NEXAVAR® tablets are round, biconvex, red film-coated tablets, debossed with the "Bayer cross" on one side and "200" on the other side.

Classification

Sorafenib is a small molecule bi-aryl urea with multikinase inhibitor activity.

Mechanism of Action

Sorafenib inhibits receptor tyrosine kinases including VEGFR2, VEGFR3, and PDGFR, receptors involved in angiogenesis. Sorafenib also inhibits the serine-threonine kinase Raf-1 in the Ras/Raf/MAPK/MEK pathway which is an important signaling pathway in cellular proliferation. Sorafenib demonstrates anti-tumor activity in hepatocellular carcinoma xenograft models, as well as specific anti-angiogenic activity with reduction in microvessel density.

Metabolism

In vitro and in vivo data indicate that sorafenib is metabolized primarily by the liver. Systemic exposure and safety data were comparable in patients with Child-Pugh A and B hepatic impairment. Sorafenib has not been studied in patients with Child-Pugh C **hepatic impairment.** No dosage adjustment is necessary when administering sorafenib to patients with Child-Pugh A and B hepatic impairment. Sorafenib has not been studied in patients undergoing renal dialysis. However, there is no evidence of increased toxicity in patients with moderate to severe **renal impairment**.

Contraindications

Sorafenib is contraindicated in patients with known severe hypersensitivity to sorafenib or any other component of NEXAVAR®.

Availability

Sorafenib is FDA-approved for the treatment of unresectable HCC and advanced RCC. Locally-obtained, commercial supplies of sorafenib will be prescribed for this study.

Storage and handling

The tablets are packaged in high-density polyethylene bottles (climate zones I/II) or alu-alu blisters (climate zones III/IV). They should only be stored in the pack provided. The storage temperature for high density polyethylene bottles should not exceed 25° C.

Refer to the sorafenib Package Insert for complete storage and handling information.

Side Effects

Complete and updated adverse event information is available in the Investigational Drug Brochure and/or product Package Insert. (Bayer 2011).

The most common adverse reactions (≥ 20%), which were considered to be related to NEXAVAR, are fatigue, weight loss, rash/desquamation, hand-foot skin reaction, alopecia, diarrhea, anorexia, nausea, and abdominal pain. Specific adverse events which may be associated with sorafenib and/or special precautions for its use include but are not limited to:

Dermatologic

Dermatological AEs (rash and HFSR) represented the most common AEs attributed to sorafenib. These events were rarely serious but did frequently lead to a transient dose interruption or reduction. Although these events may be troublesome and in some cases interfere with activities of daily living, these events were amenable to remedial therapy and were generally reversible. Management of dermatologic toxicities may include topical therapies for symptomatic relief, temporary treatment interruption and/or dose modification of sorafenib, which in severe or persistent cases, might include permanently discontinuing sorafenib. Patients with discomfort due to HFSR may be treated with topical emollients, high-potency topical steroids, or keratolytic creams (urea/salicylic acid).

Gastrointestinal

Gastrointestinal AEs (nausea, diarrhea) were the second most common toxicities associated with sorafenib. These events were rarely serious but did frequently lead to dose interruptions or reductions. The events were amenable to therapy and were generally considered an acceptable risk in this patient population. Patients with poorly tolerated GI or skin toxicities may be successfully managed with a brief dose interruption (until symptoms improve/resolve). Symptomatic therapies may also be initiated at the physician's discretion.

Gastrointestinal perforation has been reported as an uncommon event in patients taking sorafenib (reported in less than 1□ of patients taking sorafenib). In some cases this was not associated with apparent intra-abdominal tumor. Sorafenib therapy should be discontinued in case of such an event.

Hypertension

Hypertension was usually mild to moderate, occurred early in the course of treatment, and was amenable to management with standard antihypertensive therapy. Blood pressure should be checked regularly within the first 2 to 3 weeks of starting sorafenib therapy and then monitored and treated, if required, in accordance with standard medical practice. In cases of severe or persistent hypertension that persists despite antihypertensive therapy, permanent discontinuation of sorafenib should be considered.

Rare but Serious Events

Reversible posterior leukoencephalopathy, interstitial lung disease (ILD)-like events and Stevens-Johnson Syndrome have been reported as uncommon events in patients taking sorafenib.

Fatigue and Anorexia

Although fatigue and anorexia may be troublesome and in some cases severe, these events would be considered an acceptable risk in this patient population.

Vascular Events and Bleeding

The data do not suggest an appreciable risk of thrombosis or ischemic events associated with sorafenib, but in a placebo controlled study with a total of 768 patients in the intent-to-treat population, there was a slightly increased incidence of Grade 3 and 4 bleeding events in the sorafenib group; overall the incidence of Grade 3 and Grade 4 events was low (6 [1.6□] in the sorafenib group and 1 [0.3□] in the placebo group). However, there was no increase in the risk of bleeding in a population of subjects with HCC, which is normally prone to bleeding complications. Patients receiving coagulation treatment should be routinely monitored. If any bleeding event necessitates medical intervention, it is recommended that permanent discontinuation of sorafenib be considered.

There was no indication of wound healing impairment in patients undergoing surgery while taking sorafenib. There was no influence on wound healing capacity as investigated in a rat model. However, temporary interruption of sorafenib is recommended in patients undergoing

major surgical procedures. Prior to elective procedures, it is recommended that sorafenib be interrupted for 5-7 days. There is limited clinical experience regarding the timing of reinitiation of sorafenib therapy following major surgical intervention. Therefore, the decision to resume sorafenib therapy following a major surgical intervention should be based on clinical judgment taking co-morbidities and need for adequate wound healing into consideration.

Patients with vascular diseases such as wet macular degeneration, vasculitis, or new peptic ulcer or other evidence of bleeding may receive the full dose of sorafenib but require close monitoring in accordance with established medical practice.

Cardiac Ischemia; Infarction

Physicians should also discuss with patients that cardiac ischemia and/or infarction has been reported during NEXAVAR® treatment, and that they should immediately report any episodes of chest pain or other symptoms of cardiac ischemia and/or infarction.

Infections

The data suggest that sorafenib has a mild lymphopenic effect with a small increase in the risk of infection. The effect of sorafenib on leukocyte count does not pose a significant safety concern.

Hypophosphatemia

Hypophosphatemia appears to be associated with sorafenib therapy. There were no instances of severe hypophosphatemia, defined as a serum phosphate value below 1.0 mg/dL, and there were no evident clinical manifestations associated with hypophosphatemia.

Neuropathy

Sorafenib was infrequently associated with peripheral neuropathy. The timing of these events suggests an inflammatory process rather than a cumulative neurotoxic effect.

Birth Defects, Fetal Loss, and Breastfeeding

Physicians should inform female patients that sorafenib may cause birth defects or fetal loss and that they should not become pregnant during treatment with sorafenib and for at least 2 weeks after stopping treatment. Both male and female patients should be counseled to use effective birth control during treatment with sorafenib and for at least 2 weeks after stopping treatment. Female patients should also be advised against breast-feeding while receiving sorafenib.

4.2 Drug Accountability

The Investigational Pharmacist will manage drug accountability records for temsirolimus. Sorafenib will be procured from commercial supplies and accounted for according to standard of care.

4.3 Drug Ordering

Sorafenib will be ordered as a standard of care medication and will be obtained from commercial supplies.

Temsirolimus will be supplied for this trial directly by the manufacturer, Pfizer, Inc., from commercial supplies. Temsirolimus will be shipped directly to the primary study site: UCSF Helen Diller Family Comprehensive Cancer Center, and to the study sub-site: Robert H. Lurie Comprehensive Cancer Center of Northwestern University. Both sites will be responsible for direct distribution of the drug to the study patients and for drug accountability at each respective site.

To obtain temsirolimus study drug supply or for questions about temsirolimus supply, each site will contact Pfizer, Inc. directly: [redacted]

At the conclusion of the study, each institution will promptly contact the individual identified above at the research grant provider (Pfizer, Inc.) for instruction regarding whether to return all unused temsirolimus and associated diluent at the research grant provider's expense, or whether to dispose properly of temsirolimus and associated diluent at its own expense and submit documentation of such destruction. The determination as to whether the study drug shall be returned or destroyed is solely within the research grant provider's discretion.

4.4 Packaging and Labeling of Study Drugs

Drugs will be packaged and labeled per UCSF institutional standards, adhering to applicable local and federal laws.

5. Treatment Plan

5.1 Dosage and Administration

Treatment will be administered on an outpatient basis.

Temsirolimus at a dose of 10 mg weekly will be administered intravenously over 60 minutes using an infusion pump in the study site infusion center starting on Cycle 1, Day 1 of study enrollment. Infusion will be given through non-DEHP-containing tubing with an in-line polyethersulfone filter (≤ 5 microns). Premedication with diphenhydramine 25-50 mg IV will be administered 30 minutes prior to each temsirolimus infusion. Patients will be monitored by infusion center nursing staff every 15 minutes during infusion, with treatment interruption required for any evidence of an infusion reaction. If hypersensitivity symptoms develop, infusion will be stopped. Patients will be treated with additional H1 antagonist as well as an H2 antagonist (such as famotidine 20 mg or ranitidine 50 mg intravenously) at discretion of treating investigator. If hypersensitivity symptoms are mild (Grade 1 or 2) and resolve over 30-60 minutes of observation and with administration of additional H1 and H2 blocker, infusion may resume with caution at a reduced rate (over approximately 60 minutes), at the discretion of the treating investigator. Additional supportive care measures will be available in each study site infusion center in case of severe hypersensitivity reaction. Temsirolimus dosing will continue weekly (± 3 days) until unacceptable toxicity, disease progression, the withdrawal of consent by the patient, or the decision to discontinue treatment by the treating physician. Subsequent infusions starting Cycle 2, Day 1 may be given over 30 minutes if no evidence of infusion reaction is observed after the first cycle has been completed.

Patients will be instructed to take sorafenib at starting dose of 200 mg 1 tab twice daily starting on Cycle 1, Day 1 of study enrollment after completion of temsirolimus infusion. Sorafenib will be ingested with water on an empty stomach, either 1 hour before or 2 hours after food ingestion. If unable to tolerate sorafenib on an empty stomach, it may be taken with a low fat meal. The tablet will be swallowed whole. Sorafenib dosing will continue daily or twice daily without interruption until unacceptable toxicity, disease progression, the withdrawal of consent by the patient, or the decision to discontinue treatment by the treating physician (*see Section 6.3 Reasons to Withdraw a Subject*).

Table 5.1 Regimen Description

Study Drug	Premedication; precautions	Dose	Route	Schedule	Cycle Length
Temsirolimus	Premedicate with diphenhydramine 30 minutes prior to temsirolimus infusion; monitor every 15 minutes during infusion; administer with in-line filter	10 mg	IV	Weekly(± 3 days)	4 weeks (28 days)
Sorafenib	Take on empty stomach or with low fat meal	200 mg	Oral	BID	

5.2 Dose Modifications and Dosing Delays

The following adjustments should be made in the appropriate drug (depending on attribution of toxicity) based upon the guidelines below in *Section 5.2.3 Dose Modification Protocol*. If either study drug is discontinued, a patient will be removed from study.

5.2.1 Temsirolimus Dose Modifications

Patient Temsirolimus Dose Level	Temsirolimus Dose Modification
10 mg IV weekly (starting dose)	↓to 7.5 mg IV weekly
7.5 mg IV weekly	↓to 5 mg IV weekly
5 mg IV weekly	Discontinue

5.2.2 Sorafenib Dose Modifications

Patient Sorafenib Dose Level	Sorafenib Dose Modification
200 mg PO BID (starting dose)	↓ to 200 mg QD
200 mg PO QD	↓ to 200 mg PO QOD
200 mg PO QOD	Discontinue

5.2.3 Dose Modification Protocol

- Held doses of either drug will be considered as omitted. (For example, if temsirolimus is held on cycle 2, day 8, the next dose of temsirolimus the following week will be considered cycle 2, day 15, rather than delaying the cycle and repeating the day 8 dose.)
- Laboratory values should be rounded to one significant digit.
- For any toxicity (regardless of grade), despite optimal supportive care, that is felt by the treating investigator to represent a risk to the patient's safety, additional dose reduction,

treatment delay, or treatment discontinuation is permitted at the discretion of the treating investigator.

Toxicity	Grade (NCI CTCAE v4.0)	Temsirolimus Adjustment	Sorafenib Adjustment
HEMATOLOGIC			
Neutropenia	Grade 1 (ANC 1500 to < 2000/mm3)	No change	No change
	Grade 2 (ANC 1000 to < 1500/mm3)	No change	No change
	Grade 3 (ANC 500 to < 1000/mm3)	Hold both drugs and recheck weekly until ≤ Grade 2, then decrease TEM by 1 DL and continue SOR at same dose; if recurs, hold both drugs and recheck weekly until ≤ Grade 2, then decrease SOR by 1 DL and continue TEM at same dose; continue alternating dose reductions if toxicity recurs	
	Grade 4 (ANC < 500/mm3)	Hold both drugs and recheck weekly until ≤ Grade 2, then decrease both drugs by 1 DL	
Lymphopenia	Any Grade	No dose reductions or delays will be performed for lymphopenia	
Thrombocytopenia	Grade 1 (PLT 75,000 to < LLN/mm3)	No change	No change
	Grade 2 (PLT 50,000 to < 75,000/mm3)	No change	No change
	Grade 3 (PLT 25,000 to < 50,000/mm3)	Hold both drugs and recheck weekly until ≤ Grade 2, then decrease TEM by 1 DL and continue SOR at same dose; if recurs, hold both drugs and recheck weekly until ≤ Grade 2, then decrease SOR by 1 DL and continue TEM at same dose; continue alternating dose reductions if toxicity recurs	
	Grade 4 (PLT < 25,000/mm3)	Hold both drugs and recheck weekly until ≤ Grade 2, then decrease both drugs by 1 DL	
Anemia	Any Grade	No dose reductions or delays will be performed for anemia; transfusions and/or growth factor support may be used at the discretion of the investigator	
GASTROINTESTINAL (EXCLUDING HEPATIC DYSFUNCTION)			
Nausea/vomiting, constipation, or mucositis (despite optimal supportive care)	Grade 1	No change	No change
	Grade 2	No change	No change
	Grade 3	Hold both drugs until ≤ Grade 2, then decrease TEM by 1 DL and continue SOR at same dose; if recurs, hold both drugs until ≤ Grade 2, then decrease SOR by 1 DL and continue TEM at same dose; continue alternating dose reductions if toxicity recurs	
	Grade 4	Hold both drugs until ≤ Grade 2, then decrease both drugs by 1 DL	
Diarrhea (despite optimal supportive care)	Grade 1	No change	No change
	Grade 2	No change	No change
	Grade 3	Hold both drugs until ≤ Grade 2, then decrease SOR by 1 DL and continue TEM at same dose; if recurs, hold both drugs until ≤ Grade 2, then decrease TEM by 1 DL and continue SOR at same dose; continue alternating dose reductions if toxicity recurs	
	Grade 4	Hold both drugs until ≤ Grade 2, then decrease both drugs by 1 DL	

Toxicity	Grade (NCI CTCAE v4.0)	Temsirolimus Adjustment	Sorafenib Adjustment
HEPATIC DYSFUNCTION			
Hyperbilirubinemia (substitute direct bilirubin values and normal ranges for patients with Gilbert's Syndrome)	Grade 1 (BILT > ULN to 1.5 times ULN)	No change	No change
	Grade 2 (BILT > 1.5 to 3 times ULN)	Hold both drugs and recheck weekly until ≤ Grade 1, then decrease SOR by 1 DL and continue TEM at same dose; if recurs, hold both drugs and recheck weekly until ≤ Grade 2, then decrease TEM by 1 DL and continue SOR at same dose; continue alternating dose reductions if toxicity recurs	
	Grade 3 (BILT > 3 to 10 times ULN)	Hold both drugs and recheck weekly until ≤ Grade 1, then decrease both drugs by 1 DL	
	Grade 4 (BILT > 10 times ULN)	Discontinue protocol therapy	
Transaminase and alkaline phosphatase elevation (if assessed as treatment-related, clinically relevant, and ≥ 2 grades from baseline)	Grade 1 (> ULN to 2.5 times ULN)	No change	No change
	Grade 2 (> 2.5 to 5 times ULN)	No change	No change
	Grade 3 (> 5 to 20 times ULN)	Hold both drugs and recheck weekly until ≤ Grade 2, then decrease SOR by 1 DL and continue TEM at same dose; if recurs, hold both drugs and recheck weekly until ≤ Grade 2, then decrease TEM by 1 DL and continue SOR at same dose; continue alternating dose reductions if toxicity recurs	
	Grade 4 (> 20 times ULN)	Discontinue protocol therapy	
Ascites	Grade 1 (Asymptomatic)	No change	No change
	Grade 2 (Symptomatic, requiring diuretic)	No change	<u>New ascites:</u> Hold SOR and reevaluate weekly; if recovers to ≤ Grade 1, then decrease SOR by 1 DL; if recurs/worsens, decrease TEM by 1 DL; continue alternating dose reductions if toxicity recurs <u>Worsening in baseline Grade 1 or 2 ascites</u>: Optimize with supportive care including diuretic regimen as needed; if recurs/worsens, decrease SOR by 1 DL; if recurs/worsens, decrease TEM by 1 DL; continue alternating dose reductions if toxicity recurs

Toxicity	Grade (NCI CTCAE v4.0)	Temsirolimus Adjustment	Sorafenib Adjustment
	Grade 3 (Symptomatic, requiring paracentesis)	No change	New ascites: Hold SOR and reevaluate weekly; if recovers to ≤ Grade 1, then decrease SOR by 1 DL; if recurs, decrease TEM by 1 DL; if recurs/worsens despite dose reductions in both drugs plus optimal supportive care, remove from study Worsening in baseline Grade 1 or 2 ascites: Optimize with diuretic regimen and paracentesis if needed for symptoms; if repeat paracentesis is required after ≥ 2 weeks despite optimizing supportive care and diuretics, decrease sorafenib by 1 DL; if recurs/worsens, decrease TEM by 1 DL; if recurs/worsens despite dose reductions in both drugs plus optimal supportive care, remove from study
	Grade 4 (Life threatening)	Discontinue protocol therapy	

Toxicity	Grade (NCI CTCAE v4.0)	Temsirolimus Adjustment	Sorafenib Adjustment
CARDIOVASCULAR AND VASCULAR			
Hypertension	Blood Pressure (BP) ≤ 140/90	No change	
	BP >140/90 and ≤ 160/100	No change in study drugs; optimize anti-hypertensive regimen for goal BP ≤ 140/90; see section 6.1	
	BP > 160/100	If pre-treatment BP > 160/100, hold both drugs and treat according to Section 6.1; if BP remains > 160/100 **despite optimal medical management**, hold both drugs until≤ 160/100, then decrease SOR by 1 DL; if HTN recurs with BP > 160/100, repeat and decrease SOR by 1 DL; if recurs again > 160/100, discontinue protocol therapy	
	Grade 4	Discontinue protocol therapy	
Hemorrhage	Grades 1, 2 (Mild, not requiring transfusion)	No change	No change
	Grade 3 (Requiring transfusion)	No change	Hold SOR until ≤ Grade 2, HCT stable for ≥ 1 week, bleeding resolved, and there is no known anatomic or pathologic condition that significantly increases risk of recurrent bleeding, then resume at same dose; if recurs, discontinue protocol therapy
	Grade 4 (Catastrophic or CNS)	Discontinue protocol therapy	
Arterial thrombosis, including myocardial ischemia or infarction, cerebrovascular accident, or mesenteric ischemia	Any Grade	Discontinue protocol therapy	
Venous thrombo-embolism* requiring anticoagulation (*excluding portal or hepatic vessel thrombosis or tumor thrombus)	Any Grade	Discontinue protocol therapy	
RENAL AND METABOLIC			
Elevated creatinine (despite optimal supportive care, i.e. hydration if dehydrated, and if assessed as treatment-related, clinically relevant, and ≥ 1 grade from baseline)	Grade 1 (CRE > ULN to 1.5 times ULN)	No change	No change
	Grade 2 (CRE > 1.5 to 3 times ULN)	Hold both drugs and recheck weekly until ≤ Grade 1, then resume at prior doses; if recurs, hold both drugs and recheck weekly until ≤ Grade 1, then decrease TEM by 1 DL and continue SOR at same dose; if recurs again, hold both drugs and recheck weekly until ≤ Grade 1, then decrease SOR by 1 DL and continue TEM at same dose; continue alternating dose reductions if toxicity recurs	
	Grade 3 (CRE > 3 to 6 times ULN)	Hold both drugs and recheck weekly until ≤ Grade 1, then decrease both drugs by 1 DL	
	Grade 4 (CRE > 6 times ULN)	Discontinue protocol therapy	
Hypo-phosphatemia	Grade 1	No change	No change
	Grade 2	No change	No change

Toxicity	Grade (NCI CTCAE v4.0)	Temsirolimus Adjustment	Sorafenib Adjustment
(despite optimal supportive care, i.e. electrolyte repletion)	Grade 3	Asymptomatic, first occurrence: Increase oral phosphate repletion regimen up to 3.5 grams per day as tolerated, encourage increased intake of phosphorus-rich foods, and continue study drug treatment. Recheck in 1 week. If ≤ Grade 2, continue treatment at current study drug dosage with the increased phosphate repletion. If remains ≥ Grade 3, hold TEM, continue SOR, and continue increased phosphate repletion. Recheck weekly until ≤ Grade 2, then restart TEM with dose reduction by 1 DL and continue SOR at prior dose. Asymptomatic, recurrent: If recurs after 1 dose level reduction in TEM despite continuing the highest tolerable dose of oral phosphate supplementation*, hold both drugs, continue repleting phosphate, and recheck weekly. Once recovers to ≤ Grade 2, decrease SOR by 1 DL and continue TEM at prior dose. Continue this protocol with alternating drug dose reductions if asymptomatic Grade 3 toxicity recurs. *If asymptomatic Grade 3 hypophosphatemia recurs but patients **have not been taking the highest tolerable dose of oral phosphate supplementation**, follow the hypophosphatemia algorithm above as for the first occurrence of asymptomatic grade 3 hypophosphatemia. Symptomatic, first occurrence: Increase oral phosphate repletion regimen up to 3.5 grams per day as tolerated, encourage increased intake of phosphorus-rich foods, and hold both TEM and SOR. Consider cautious intravenous phosphate supplementation at the investigator's discretion and according to standard institutional protocols. Recheck weekly until ≤ Grade 2 and asymptomatic, then decrease TEM by 1 DL and resume SOR at prior dose. Symptomatic, recurrent: If recurs after 1 DL reduction in TEM and despite continuing the highest tolerable dose of oral phosphate supplementation*, hold both drugs and continue repleting phosphate. Consider cautious intravenous phosphate supplementation at the investigator's discretion and according to standard institutional protocols. Recheck weekly. Once recovers to ≤ Grade 2 and asymptomatic, decrease SOR by 1 DL and continue TEM at prior dose. Continue this protocol with alternating drug dose reductions if toxicity recurs. *If symptomatic Grade 3 hypophosphatemia recurs but patients **have not been taking the highest tolerable dose of oral phosphate supplementation**, follow the hypophosphatemia algorithm above as for the first occurrence of symptomatic grade 3 hypophosphatemia.	
	Grade 4 **(< 1.0 mg/dL)**	Follow protocol above for **symptomatic** Grade 3 hypophosphatemia.	
Hyperglycemia	Grade 1	No change	No change

Toxicity	Grade (NCI CTCAE v4.0)	Temsirolimus Adjustment	Sorafenib Adjustment
(despite optimal supportive care)	Grade 2	No change	No change
	Grade 3	Hold TEM and recheck weekly until ≤ Grade 2, then decrease by 1 DL	No change
	Grade 4	Discontinue protocol therapy	
Hyperlipidemia (despite optimal supportive care)	Grade 1	No change	No change
	Grade 2	No change	No change
	Grade 3	Hold TEM and recheck weekly until ≤ Grade 2, then decrease by 1 DL	No change
	Grade 4	Hold TEM and recheck weekly until ≤ Grade 2, then decrease by 1 DL	No change
DERMATOLOGIC			
Hand-foot syndrome (despite optimal supportive care)	Grade 1 (Numbness, discomfort, dysesthesia/paresthesia, tingling, painless swelling, or erythema that does not disrupt normal activities)	No change	No change
	Grade 2 (Painful erythema, swelling, or discomfort that disrupts normal activities)	No change	Hold SOR until ≤ Grade 1, then resume at prior dose; if recurs, hold SOR until ≤ Grade 1, then decrease by 1 DL
	Grade 3 (Moist desquamation, ulceration, blistering, or severe pain that disrupts ADL)	No change	Hold SOR until ≤ Grade 1, then decrease by 1 DL
	Grade 4	Discontinue protocol therapy	
Other dermatologic toxicity (including dermatitis, non-healing wound, pruritis, desquamation) (despite optimal supportive care)	Grade 1	No change	No change
	Grade 2	No change	Hold SOR until ≤ Grade 1, then resume at prior dose; if recurs, hold SOR until ≤ Grade 1, then decrease by 1 DL
	Grade 3	Hold both drugs until ≤ Grade 1, then reduce SOR and/or TEM by 1 DL based upon the investigator's attribution of toxicity	
	Grade 4	Discontinue protocol therapy if severe or life-threatening toxicity, versus apply modification for Grade 3	
OTHER			
Other toxicities* (despite optimal supportive care)	Grade 1	No change	No change
	Grade 2	No change	No change
	Grade 3	Hold both drugs until ≤ Grade 1, then reduce SOR and/or TEM by 1 DL based upon the investigator's attribution of toxicity	
	Grade 4	Discontinue protocol therapy if severe or life-threatening toxicity, versus apply modification for Grade 3	

*For any toxicity (regardless of grade), despite optimal supportive care, that is felt by the treating investigator to represent a risk to the patient's safety, additional dose reduction, treatment delay, or treatment discontinuation is permitted at the discretion of the treating investigator.

5.2.4 Unacceptable Toxicities

Patients must be removed from the study and protocol therapy discontinued for toxicities deemed by the treating investigator as unacceptable. These toxicities include but are not limited to:

- Thrombotic events, including cerebrovascular accident, myocardial infarction or ischemia, gastrointestinal infarction or ischemia, pulmonary embolus, or any thrombotic event requiring anticoagulation. New appearance of hepatic or portal vein thromboses are excluded. Patients with superficial thrombophlebitis, superficial venous thrombosis, or portal venous or hepatic vessel thrombosis not requiring anticoagulation may remain on therapy at the discretion of the treating physician.
- Recurrent grade 3 or any grade 4 hemorrhage.
- Gastrointestinal perforation requiring medical or surgical therapy.
- Wound dehiscence requiring medical or surgical therapy.
- Grade 4 hypertension despite optimal medical management.
- Grade 4 hypersensitivity reaction to either study drug, or Grade 3 reaction that recurs despite optimal supportive care.
- Any Grade 3 or 4 toxicity that is deemed serious and/or life-threatening by the treating investigator.
- Any irreversible (≥ 2 weeks despite optimal supportive care) Grade 3 or 4 toxicity (excluding nausea, vomiting, asymptomatic Grade 3 hypophosphatemia or Grade 3 hyponatremia, anemia, lymphopenia, or fatigue).

5.3 Management of Toxicities

Each patient receiving at least 1 dose of temsirolimus in combination with at least 1 dose of sorafenib will be evaluable for safety. The safety parameters include all laboratory tests and hematological abnormalities, physical findings, and spontaneous reports of adverse events reported to the investigator by patients.

Each patient will be assessed periodically for the development of any toxicity as outlined in Table 6.1, Schedule of Study Procedures and Assessments. Toxicity will be assessed according to the NCI CTCAE v4.0. Dose adjustments, treatment delay, and treatment discontinuation will be determined according to the system showing the greatest degree of toxicity. Toxicity will be formally assessed at protocol-defined assessment timepoints well as by investigator review of weekly laboratory testing and, in the interim, by patient contact with the study coordinator and/or treating investigator should new symptoms arise between visits. General principles of toxicity management are listed below:

- Temsirolimus and sorafenib will be dose-adjusted according to investigator's attribution of toxicity. In equivocal cases, the decision may be made by treating investigator.
- Asymptomatic changes in laboratory values require dose modification only if assessed to be treatment-related, clinically relevant, and represent a change of ≥ 2 grades from baseline.
- If toxicity occurs, supportive care medications should be applied to ameliorate signs and symptoms (including antiemetics for nausea and vomiting, antidiarrheals for diarrhea, antipyretics and antihistamines for drug fever, antihypertensives for hypertension, and phosphate repletion for hypophosphatemia) before toxicity grade is determined.

- A patient will be removed from protocol therapy if either of the two study drugs is discontinued.

- Temsirolimus will be discontinued if it is not tolerated at a dose of 5 mg IV weekly.
- Sorafenib will be discontinued if it is not tolerated at a dose of 200 mg PO every other day.
- Once a dose has been reduced for toxicity, it should not be re-escalated.
- In obese patients, all dosing is to be determined by the patient's actual body weight.
- If there is more than one indication for dose modification, use the more strict dose modification (i.e. modify according to the most severe toxicity).
- If treatment with either of the two study drugs is held for treatment-related toxicity for longer than 28 days (1 cycle), a patient should be removed from study.
- In selected cases where the investigator assesses the patient as having clinical benefit from protocol therapy, treatment may be resumed after > 28 day delay along with dose reduction, optimal supportive care, and close monitoring at the discretion of the treating investigator with the approval of the Study Chair.

5.3.1 Supportive Care Guidelines

Supportive medications to alleviate or mitigate adverse effects of protocol therapy may include the following agents at the discretion of the treating investigator and according to institutional standards of care. All medications must be reviewed for potential interaction with study drug therapy (see Appendices). The treating investigator, Investigational Pharmacist, and/or Study Chair should be alerted if the patient is prescribed any agent known to affect or with the potential to affect selected P450 isoenzymes or other drug metabolism pathways.

- Antidiarrheal Agents: Patients should be instructed to begin loperamide at the earliest signs of a poorly formed or loose stool. Oral loperamide should be prescribed in the following manner: 4 mg at first onset of diarrhea then 2 mg every 2 hours until diarrhea-free for at least 12 hours up to 16 mg daily.
- Antiemetic Agents: Drugs such as lorazepam, prochlorperazine, or serotonin antagonists may be used if clinically indicated.
- Antihistamines: An antihistamine such as diphenhydramine may be used to manage dermatitis associated with therapy at the discretion of the treating physician.
- Anti-diabetic Agents: For patients who develop clinically significant hyperglycemia on study protocol, anti-diabetic therapy and dietary modification are required. The oral anti-diabetic agents, glyburide or metformin, may be instituted at the discretion of the treating physician, assuming no contraindication. Metformin must be temporarily discontinued for ≥ 48 hours before and after iodinated contrast imaging, with confirmation of stable renal function prior to resuming this agent after contrast exposure. Use of glipizide and glimepiride (CYP2C9 substrates) should be avoided or used with caution due to potential interaction with sorafenib. Insulin may be instituted at the discretion of the treating physician to optimize glycemic control. Management of complications of hyperglycemia should be performed by treating physician according to institutional standard practices. All patients who develop clinically significant hyperglycemia on study protocol must receive diabetic teaching and education materials.
- Antihyperlipidemic Agents: Patients who develop serum cholesterol > 350 mg/dL and/or triglycerides > 300 mg/dL while on therapy will require medication (assuming no contraindications and with standard precautions as appropriate for the class of agents chosen) and/or dietary modifications at the discretion of the treating investigator. Pravastatin or rosuvastatin is recommended if LDL and/or total cholesterol are elevated, with or without

elevated triglycerides, because these agents are not CYP 3A4 substrates. Atorvastatin may also be used with caution. If tryglycerides alone are elevated, fenofibrate may be considered.

- Antihypertensive Agents: Patients who develop blood pressures > 140/90 mm Hg on at least 2 occasions should be managed with oral antihypertensives including amlodipine, hydrochlorothiazide, or metoprolol by their treating physician. Angiotensin converting enzyme (ACE) inhibitors should be used with caution due to increased risk for angioedema when used in combination with temsirolimus. Use of angiotensin receptor blockers (ARB) which are CYP2C9 substrates as well as inhibitors should be avoided or used with caution due to potential for interaction with sorafenib which is also a CYP2C9 inhibitor. Blood pressure and any relevant laboratory parameters should be rechecked within 7 days of starting a new medication.
- Diuretics: Diuretics such as furosemide may be used for the management of periorbital or lower extremity edema or ascites at the discretion of the treating physician. Severe episodes of edema, such as congestive heart failure, pleural effusion, ascites, pericardial effusion, or pulmonary edema may require additional measures such as percutaneous drainage or hospitalization.
- Electrolyte Supplementation: Electrolytes should be repleted according to standard institutional protocols. Specific recommendations for prevention and management of **hypophosphatemia,** a common side effect of temsirolimus, are provided below:
  - For patients enrolled on study with baseline hypophosphatemia ≥ Grade 2 (< 2.5 mg/dL) prior to start of study drug therapy, and for patients who develop hypophosphatemia on study drug treatment, phosphate supplementation should be initiated with over-the-counter phosphate tablets, caplets, or powders. Recommended starting dosage is 250 mg to 500 mg elemental phosphate four times daily, assuming no contraindications (such as hyperkalemia, severe diarrhea or dehydration, or severe renal insufficiency). The dose may be increased up to up to a total dose of 3.5 grams daily, assuming no contraindications. Administer after meals and at bedtime. Serum phosphate should be rechecked within 7 days after starting supplements.
  - For patients on study drug therapy who have had prior episodes of hypophosphatemia, oral phosphate supplementation should be continued until serum phosphorus level is ≥ 3.0 mg/dL. Supplementation should be restarted if level < 2.5 mg/dL.
  - For patients who have discontinued study drug therapy, phosphate supplementation and monitoring for purposes of the study may be discontinued once serum phosphate level returns to baseline.
  - Phosphate levels should be rechecked within 7 days after starting supplements, and followed at least weekly thereafter while on protocol therapy.
  - Intravenous therapy may be administered at the treating investigator's discretion. Intravenous repletion should be administered with caution according to standard institutional protocols.
  - All patients enrolled in study should be provided list of phosphorus-rich foods. Patients with serum phosphate levels < 2.5 mg/dL should be encouraged to increase intake of these foods if tolerated and no contraindications.
- Growth Factors: Erythropoietin or darbopoietin are permitted at the discretion of the treating physician. Filgrastim and PEG-filgrastim (Neulasta) should not be used in place of protocol-specified dose reductions or delays for myelosuppression, nor should they be used prophylactically because of concern for myelosuppression from a prior cycle of chemotherapy. For treatment of febrile neutropenia, the use of colony-stimulating factors should not be routinely instituted as an adjunct to appropriate antibiotic therapy. The use of granulocyte colony-stimulating factors may be indicated, however, in subjects who have prognostic factors

that are predictive of clinical deterioration such as pneumonia, hypotension, multi-organ dysfunction, sepsis syndrome, or fungal infection. Investigators should use discretion in initiating colony-stimulating factors in these settings.

- Topical Agents: Topical steroid creams may be used to manage pruritic skin toxicity at the discretion of the treating physician. Non-alcohol-based topical emollients such as Aquaphor may be used for hand-foot syndrome or desquamating rashes. Non-alcohol-based moisturizers may be used for dry skin.
- Treatment of Infusion Reactions: Subjects who experience study drug-associated temperature elevations ≥ 38.5°C or other infusion-related symptoms may be treated symptomatically with acetaminophen, diphenhydramine, meperidine, or other medications at the discretion of the treating investigator. See Section 4.1.1 and Package Insert and Investigator's Brochure for temsirolimus for further information on management of infusion reactions.(Pfizer 2011; Wyeth 2011)

6. Study Procedures and Observations

6.1 Schedule of Procedures and Observations

The study-specific assessments are detailed in this section and outlined in *Table 6.1 Schedule of Study Procedures and Assessments*. For more details on the study procedures, please refer to Study Laboratory and Procedures Manual.

Screening assessments must be performed within 28 days prior to the first dose of investigational product.

All on-study visit procedures are allowed a window of ± 7 days unless otherwise noted.

All patients who are consented will be registered in OnCore®, the UCSF Helen Diller Family Comprehensive Cancer Center Clinical Trial Management System (CTMS). The system is password protected and meets HIPAA requirements. A written, signed, informed consent form (ICF) and a Health Insurance Portability and Accountability Act (HIPAA) authorization must be obtained before any study-specific assessments are initiated. A copy of the signed ICF will be given to the subject and a copy will be filed in the medical record. The original will be kept on file with the study records.

6.1.1 Pretreatment Period

Patients with hypertension, diabetes mellitus, hyperlipidemia, and/or hepatitis B virus infection require optimization of these conditions prior to starting protocol therapy if found eligible upon screening.

6.1.1.1 Screening Assessments

The Screening procedures and assessments must be completed within 28 days of the Day 1 Visit unless otherwise stated. Any results falling outside of the reference ranges may be repeated at the discretion of the investigator.

- Complete physical examination, including height and weight measurement
- Vital signs, including temperature, blood pressure, heart rate, pain score, and oxygen saturation
- Complete medical history with annotation to include demographics, past medical history, presentation at diagnosis, prior biopsy pathologic results (if any), prior radiologic assessments, prior therapies (including surgery, radiation, or local therapies such as chemoembolization), response to prior therapies, and review of systems

- Documentation of disease status including CT chest (with or without contrast) and contrast-enhanced CT and/or MRI abdomen/pelvis, preferably multiphasic
- ECOG Performance Status – see Appendix 1
- Child-Pugh class assessment – see Appendix 2
- BCLC and CLIP stage – see Appendix 3
- History of prior treatments and any residual toxicity relating to prior treatment
- Baseline medications taken within 28 days of Day 1
- Obtain archival tumor specimens (prefer at least 1 tumor block and/or at least 10 unstained slides plus 1 H&E slide) and pathology report (locally-obtained pathology report is acceptable at start of study) to confirm diagnosis of HCC, and for specimen banking of left-over frozen or fixed tumor specimens from all enrolled patients; patients may start study prior to obtaining the archival specimen(s) provided the pathology report has been reviewed and filed in study records
- Screening laboratory parameters: CBC w/ Diff, complete metabolic panel (electrolytes, BUN, creatinine, glucose, calcium, magnesium, phosphorus, total bilirubin, alkaline phosphatase, albumin, AST, and ALT), PT/INR, fasting blood glucose, hemoglobin A1c, fasting total cholesterol and triglycerides; hepatitis B and C serologies (HBsAb, HBsAg, HBcAb total and IgM; HCV Ab) (hepatitis B and C serologies may be performed >28 days from Day 1)
- Measurement of hepatitis B viral load (HBV DNA quantitative by PCR) if HBsAg, HBcAb total, and/or HBcAb IgM is/are positive (within 28 days of Day 1)
- Confirmation of antiviral therapy with an appropriate antiviral agent for HBV is required in patients with positive hepatitis B surface antigen, HBcAb IgM, and/or viral load - appropriate first line agents include entecavir, tenofovir, and lamivudine (note that lamivudine has higher resistance rates)
- Patients with a positive HBV viral load, HBcAb IgM, and/or HBsAg should be referred to a hepatologist if not already under the care of a hepatologist
- Initiate standing phosphate repletion for patients with baseline ≥ Grade 2 hypophosphatemia – see *Section 5.3.1 Dose Modification and Dosing Delays*
- Confirmation of adequate blood pressure control with agents compatible with study drugs – *see Sections 5.3.1 and 6.7* and *Appendices 6-9 Prohibited Medications*
- Serum or urine pregnancy test for women of childbearing potential within 14 days from start of study prior to the start of study drug

6.1.2 Treatment Period

6.1.2.1 Study Procedures – Cycle 1

Days 1, 8, 15, and 22 (within 7 days)

- Complete physical examination, including height and weight measurement
- Vital signs, including temperature, blood pressure, heart rate, pain score, and oxygen saturation
- Evaluation of clinical response or deterioration
- Evaluation of adverse events
- Concomitant medications

- Safety laboratory assessments: CBC w/ Diff, complete metabolic panel (electrolytes, BUN, creatinine, glucose, calcium, magnesium, phosphorus, total bilirubin, direct bilirubin, alkaline phosphatase, albumin, AST, and ALT), INR
- Serum AFP tumor marker (Day 1 only)
- Blood specimen collection for CTC testing (Day 1 only)
- Banking of blood specimens for possible future correlative studies (Day 1 and 8 only) (See Study Laboratory and Procedures Manual)

6.1.2.2 Study Procedures – Day 1 of Cycles 2 and beyond (within 7 days)

- Complete physical examination, including height and weight measurement
- Vital signs, including temperature, blood pressure, heart rate, pain score, and oxygen saturation
- Evaluation of clinical response or deterioration
- Evaluation of adverse events
- Concomitant medications
- ECOG Performance Status
- Safety laboratory assessments: CBC w/ Diff, complete metabolic panel (electrolytes, BUN, creatinine, glucose, calcium, magnesium, phosphorus, total bilirubin, direct bilirubin, alkaline phosphatase, albumin, AST, and ALT), INR
- Fasting total cholesterol, triglycerides, and blood glucose (odd cycles only)
- Hepatitis B viral DNA quantitative in patients with positive hepatitis B viral load at baseline and/or positive HBsAg, HBcAb total, and/or HBcAb IgM (odd cycles only or more frequently at investigator's discretion).
- Imaging with CT chest (with or without contrast) plus triphasic CT abdomen and pelvis with intravenous contrast or triphasic gadolinium-enhanced MRI abdomen/pelvis to assess sites of known tumor using RECIST 1.1 measurements and to monitor for lung toxicity (odd cycles only, every 8 weeks ± 7 days, starting Cycle 3, Day 1)
- Initial assessment of response or progression will be made based upon standard of care Radiology review plus clinical assessment if RECIST 1.1 measurements are not available at the time of study visit. In patients without signs of clinical progression, progression must be confirmed by RECIST 1.1 prior to removal from study. If progression is identified by RECIST 1.1 that was not identified by standard of care Radiology review plus clinical assessment, a patient will be removed from study at date of next scheduled study visit. Efforts will be made to obtain RECIST 1.1 measurements for patients treated at both sites within approximately 7 days of start of odd cycles.
- Serum AFP tumor marker
- Blood specimen collection for CTC testing (Cycles 2 and 3 only)
- Banking of blood specimens (Cycles 3 and End-of-Treatment only)

6.1.3 6.1.2.3 Study Procedures Day 8, 15, and 22 of All Cycles after Cycle 1 (within 7 days)

- Weight measurement and vital signs including temperature, blood pressure, heart rate, pain score, and oxygen saturation
- Safety laboratory assessments: CBC w/Diff, complete metabolic panel (electrolytes, BUN, creatinine, glucose, calcium, magnesium, phosphorus, total bilirubin, direct bilirubin, alkaline phosphatase, albumin, AST, and ALT), INR

6.1.4 End-of-Treatment Study Procedures (within 60 days)

End-of-Treatment is defined as the day on which a decision is made by an investigator to discontinue a patient's participation in the study, often due to disease progression or toxicity, or by a patient to withdraw consent; this may also be referred to as the last day on study and may not necessarily coincide with the last day of protocol therapy, particularly if study drug has been held for toxicity.

- Complete physical examination, including height and weight measurement
- Vital signs, including temperature, blood pressure, heart rate, pain score, and oxygen saturation
- Update medical history
- Evaluation of clinical response or deterioration
- Evaluation of adverse events
- Concomitant medications
- ECOG Performance Status
- Child-Pugh class assessment
- Safety laboratory assessments: CBC w/ Diff, complete metabolic panel (electrolytes, BUN, creatinine, glucose, calcium, magnesium, phosphorus, total bilirubin, direct bilirubin, alkaline phosphatase, albumin, AST, and ALT), INR; serum AFP and HBV DNA quantitative by PCR should be measured if not done within 4 weeks
- If not done within preceding 8 weeks, imaging with CT chest (with or without contrast) plus triphasic CT abdomen and pelvis with intravenous contrast or triphasic gadolinium-enhanced MRI abdomen/pelvis to assess sites of known tumor using RECIST 1.1 measurements and to monitor for lung toxicity
- Banking of blood specimens if >4 weeks since specimens collected for banking

6.1.5 Post-Treatment/Follow Up Visits

The study termination visit will be the last follow-up assessment visit for patients removed from study for disease progression with < grade 2 treatment-related toxicity and stable or undetectable hepatitis B load, if applicable. Such patients will follow up with their primary oncologist and hepatologist (if applicable) after removal from study.

Patients who are experiencing clinically relevant, treatment-related ≥ grade 2 toxicities or other adverse events at study termination will be followed periodically at intervals determined by the treating investigator until resolution, stabilization, or the assessment of irreversibility of the toxicity or toxicities in question, until the patient starts a new treatment, or until death, whichever comes first. For these patients, the study coordinator and treating investigator will confer within 8 weeks after a patient's study termination, to determine the resolution, stabilization, irreversibility and/or need for further follow-up of ongoing, clinically relevant, treatment-related ≥ grade 2 toxicities present at study termination, based upon clinical encounter, telephone follow-up, and/or medical records.

Follow up for ongoing treatment-related grade ≥ 2 toxicities at the time of study termination may also be performed at an outside physician's office if deemed in the patient's best interest, provided that the results are faxed as source documents to the treating investigator and site clinical research coordinator. (For example, follow up for thrombocytopenia on study drugs may be performed at an outside laboratory and faxed to the study site. Examination to confirm resolution of mucositis or hand-foot syndrome off study may be performed by an outside physician provided that a copy of the encounter note documenting the physical examination findings is filed in the patient's study chart. The treating investigator may also confirm resolution of symptomatic toxicities, such as hand-foot syndrome or stomatitis, with the patient by telephone.)

6.1.6 Long Term/Survival Follow-up Procedures

Patients will be followed every 3-6 months (±3 months) for up to 5 years after study discontinuation or until death, whichever comes first.

The following procedures will be performed at standard-of-care clinic visits with treating investigator or by telephone calls from treating investigator or a designated study coordinator (± 2 months):

- Evaluation of clinical response or deterioration.
- Overall survival will be measured from time of first dose of protocol therapy to date of death; for patients who are lost to follow up, survival will be censored at date last known to be alive.
- Evaluation of any residual symptomatic adverse events (if applicable).
- Other anti-cancer therapies.

Table 6.1 Schedule of Study Procedures and Assessments

Period/Procedure	Screening	Cycle 1				Cycle 2 and future Cycles				End of Treatment visit	Follow Up Visit*
Study Day/Visit Day	**-28 to 1**	**1**	**8**	**15**	**22**	**1**	**8**	**15**	**22**		
Informed consent	X										
Archival Specimen Collection [1]	X										
AE assessment		X	X	X	X	X				X	X
Concomitant medications	X [2]	X	X	X	X	X				X	
Treatment/Drug Administration											
Temsirolimus[17]		X	X	X	X	X	X	X	X		
Sorafenib [3]		X	X	X	X	X	X	X	X		
Clinical procedures											
Physical exam	X	X	X	X	X	X				X	
Vital signs	X	X	X	X	X	X	X [4]	X	X	X	
Medical history	X									X	X [6]
Liver disease assessment [5]	X										
Performance Status	X					X				X	
Laboratory procedures											
Hematology (CBC w/Diff)	X	X	X	X	X	X	X	X	X	X	
Blood chemistry [7]	X	X	X	X	X	X	X	X	X	X	
Coagulation [8]	X	X	X	X	X	X	X	X	X	X	
AFP		X				X				X	
HCV, HBV sAb, HBV sAg, HBV cAb total & IgM[16]	X										
Hemoglobin A1c	X										
Hepatitis B viral DNA quantitative by PCR [9]	X[9]					X [9]				X[9]	
Fasting total cholesterol, triglycerides, and blood glucose[10]	X[10]					X[10]					

Table 6.1 Schedule of Study Procedures and Assessments

Period/Procedure	Screening	Cycle 1				Cycle 2 and future Cycles				End of Treatment visit	Follow Up Visit*
Study Day/Visit Day	**-28 to 1**	**1**	**8**	**15**	**22**	**1**	**8**	**15**	**22**		
Standing phosphate repletion [11]	X[11]	X[11]				X[11]					
CTC [12]		X[12]				X[12]					
Banking blood samples[13]		X[13]	X[13]			X[13]				X [13]	
Pregnancy test (HCG)	X										
Imaging procedures											
Imaging (CT or MRI)	X					X [14]				X [15]	

* Patients will be followed every 3-6 months for up to 5 years after study discontinuation or until death, at standard-of-care clinic visits with treating investigator or by telephone calls from treating investigator or a designated study coordinator (± 3 months)

[1] May start study prior to obtaining specimens if pathology report is available

[2] Confirmation of antiviral therapy with an appropriate antiviral agent for HBV is required in patients with positive hepatitis B surface antigen, HBcAb IgM, and/or viral load

[3] Sorafenib will be taken orally twice daily at starting dose with modifications per Section 5.2.3

[4] Weight and vital signs on Days 8, 15, 22 of Cycle 2 and future cycles

[5] Disease assessment including Child-Pugh, BCLC, and CLIP scores

[6] Evaluation of clinical response or deterioration at Follow-Up Visit: overall survival will be measured from time of first dose of therapy to date of death, evaluation of any residual symptomatic adverse events (if applicable), other anti-cancer therapies

[7] Complete metabolic panel (electrolytes, BUN, creatinine, glucose, calcium, magnesium, phosphorus, total and direct bilirubin, alkaline phosphatase, albumin, AST, and ALT)

[8] Prothrombin time/ international normalized ratio (PT/INR)

[9] Hepatitis B viral DNA quantitative in patients with positive hepatitis B viral load at baseline (Screening) and/or positive HBsAg, HBcAb total, and/or HBcAb IgM (odd cycles only or more frequently at investigator's discretion), and end of treatment if not measured within 4 weeks prior.

[10] Screening and Day 1 of odd cycles only (Cycle 3, 5, 7, etc.)

[11] Initiate standing phosphate repletion for patients with baseline ≥ Grade 2 hypophosphatemia; adjust dosage as clinically indicated at each visit

[12] CTC on Day 1 of Cycles 1, 2 & 3 only. See Study Laboratory and Procedures Manual for instructions

[13] Banking blood samples on Cycle 1 Day 1, Cycle 1 Day 8, and Cycle 3 Day 1, and at End of Treatment (if not done within 4 wks). See Study Laboratory and Procedures Manual for instructions

[14] Odd cycles only, every 8 weeks ± 7 days, starting Cycle 3, Day 1

[15] If not done within 8 weeks, imaging with CT chest (with or without contrast) plus triphasic CT abdomen and pelvis with intravenous contrast or triphasic gadolinium-enhanced MRI abdomen/pelvis

[16] May use values from >28 days from registration

[17] Temsirolimus infusion may be performed within ± 3 days

6.2 Usage of Concurrent/Concomitant Medications

Concomitant medications are any prescription medications or over-the-counter preparations used by the patient within 28 days of initiating study treatment, during study treatment, or until 28 days following study termination. The details of all concurrent medications including vitamins and alternative therapies will be recorded on the case report forms (CRFs). All medications must be reviewed for potential interaction with study drug therapy. All concomitant medications and blood products, as well as supportive interventions (such as analgesic use, paracentesis, etc.) received by patients from screening until the end of study visit will be recorded in CRFs. Because there is a potential for interaction between the study drugs temsirolimus and sorafenib and other concomitantly administered drugs through hepatic metabolism by the cytochrome P450 system (predominantly CYP3A4), P-glycoprotein metabolism, and/or through glucuronidation by UGT1A1 and UGT1A9, the case report form must capture the concurrent use of all other drugs, over-the-counter medications, or alternative therapies. The Study Chair should be alerted if the patient is taking any agent known to affect or with the potential to affect selected P450 isoenzymes or other drug metabolism pathways.

Use of any inducers and strong inhibitors of CYP3A4 is prohibited while undergoing protocol therapy. Use of substrates of CYP3A4, CYP2B6, CYP2C8, and CYP2C9 with a narrow therapeutic range or which are assessed as sensitive by FDA guidelines is also prohibited.(2009) Please refer to Appendices B6-9 for definitions and lists of these agents. Patients who have taken medications that are known to be inducers or strong inhibitors of CYP3A4 within 28 days before registration will be excluded from study.

Substrates of CYP3A4, CYP2D6, CYP2C8, and CYP2C9 with a narrow therapeutic range or assessed as sensitive by FDA guidelines must not have been taken within 5 half-lives of the specific drug in question at the time of start of study. Medications which are weak to moderate inhibitors of CYP3A4 enzymes must be reviewed by the Study Chair or Investigational Pharmacist for approval before concomitant use. Inhibitors or inducers of P-gp, substrates of UGT1A1 and UGT1A9, and other substrates of CYP3A4, CYP2B6, CYP2C8, and CYP2C9 should be avoided or used with caution. Selected medications and substances known or with the potential to interact with or be metabolized by relevant cytochrome P450 isoenzymes (CYP3A4, CYP2B6, CYP2C8, CYP2C9), P-glycoprotein pathway, and/or UGT1A1 and UGT1A9 glucuronidation are provided in Appendices 6-9. If the investigator is unsure of potential for drug-drug interaction, the Investigational Pharmacist will be consulted. For the most up to date table of cytochrome P450-related drug interactions, please refer to the Indiana University drug interaction table at the following website:

http://medicine.iupui.edu/clinpharm/ddis/ClinicalTable.asp

6.3 Reasons to Withdraw a Subject

The Investigator will withdraw a patient whenever continued participation is no longer in the patient's best interests. Reasons for withdrawing a patient include, but are not limited to, disease progression, the occurrence of an adverse event or a concurrent illness, a patient's request to end participation, or simply significant uncertainty on the part of the Investigator that continued participation is prudent. There may also be administrative reasons to terminate participation, such as concern about a patient's compliance with the prescribed treatment regimen.

6.4 Stopping Rules

No interim assessment for primary endpoint is planned, but there will be a planned interim safety analysis by the UCSF Data and Safety Monitoring Committee after the first 30% of the planned study participants (8 patients) have enrolled and completed at least one cycle of protocol therapy or been removed for toxicity. Study accrual will not be interrupted for the early

safety assessment. The proportion of patients with treatment-related adverse events of grade 3 or higher and/or an unacceptable toxicity event (with exceptions defined below) will be summarized with 90% two-sided exact confidence intervals. Each confidence interval will be no wider than ±0.17. If the lower confidence bound (LCB) exceeds 0.17, adverse events will be evaluated and a decision will be made whether to amend the protocol or close the study.

If the true rate of treatment-related toxicity is 25%, the probability that 3 or more patients will have adverse events of grade 3 or greater is 32%. A summary of probabilities for a range of true toxicity rates is shown below. If 3 or more patients among the first 8 experience an eligible grade 3 or higher and/or unacceptable toxicity event, a decision will be made whether to amend the study or close the study due to toxicity.

True Toxicity Rate	***Probability 3 or more of 8 patients experience AEs of grade 3 or greater***	***Probability 2 or more of 8 patients experience AEs of grade 3 or greater***	***Probability 1 or more of 8 patients experience AEs of grade 3 or greater***
10%	3.8%	18.7%	57%
20%	20.3%	50.0%	83%
25%	32.0%	63.3%	89%
30%	44.8%	74.5%	94%

Exceptions of toxicity which will not be included for interim safety analyses are:

- Asymptomatic Grade 3 toxicity in laboratory values despite optimal supportive care (such as hydration or electrolyte repletion) must be treatment-related, clinically relevant*, and represent a change of ≥ 2 grades from baseline. Grade 3 hypertriglyceridemia is excluded if peak serum triglycerides are < 1500 mg/dL and recover to < 300 mg/dL with optimal medical management by the start of Cycle 2. Asymptomatic Grade 3 hypophosphatemia is also excluded.
- Grade 3 hypertension that resolves to <140/90 within 14 days of adjustment or addition of oral blood pressure medication(s) is also excluded. Hypertension that requires hospitalization or intravenous medication for control will be considered unacceptable toxicity.
- Grade 3 ascites, encephalopathy, or other toxicities associated with chronic liver disease must be ≥ 2 grades greater than baseline to be considered a DLT.
- Fatigue is excluded.
- Toxicity attributed to hepatocellular carcinoma is excluded.

*Clinically relevant is defined as toxicity which places the patient at risk for morbidity by the judgment of the treating investigator.

6.5 Medical Guidelines for the Treatment of an Overdose

There is no specific treatment for TORISEL® intravenous overdose. Temsirolimus has been administered to patients with cancer in phase 1 and 2 trials with repeated intravenous doses as high as 220 mg/m2. The risk of several serious adverse events, including thrombosis, bowel

perforation, interstitial lung disease (ILD), seizure, and psychosis, is increased with doses of temsirlimus greater than 25 mg.(Pfizer 2011; Wyeth 2011)

There is no specific treatment for NEXAVAR overdose. The highest dose of sorafenib studied clinically is 800 mg twice daily. The adverse reactions observed at this dose were primarily diarrhea and dermatologic. No information is available on symptoms of acute overdose in animals because of the saturation of absorption in oral acute toxicity studies conducted in animals. In cases of suspected overdose, sorafenib should be withheld and supportive care instituted.(Bayer 2011)

6.6 Dietary Restrictions

Avoid grapefruit and grapefruit juice (may increase levels or toxicity of sirolimus).

Avoid supplements containing St. John's Wort, alfalfa, aloe, bilberry, bitter melon, burdock, celery, damiana, echinacea, fenugreek, garcinia, garlic, ginseng (American), gymnema, marshmallow, stinging nettle.

6.7 Prohibited Medications

6.7.1 Temsirolimus

Temsirolimus is metabolized by CYP3A4 to its active metabolite sirolimus which is a substrate of both CYP3A4 and P-glycoprotein. Temsirolimus also inhibits P-glycoprotein. Temsirolimus has been shown to inhibit CYP2D6 *in vitro*, but no effect is expected *in vivo*. See Appendices 4-7.

CYP3A4 inhibitors

May decrease metabolism of temsirolimus and sirolimus. Strong inhibitors of CYP3A4 are prohibited; other inhibitors should be avoided or used with caution.

CYP3A4 Inducers

May increase metabolism of temsirolimus and sirolimus. All inducers of CYP3A4 are prohibited.

Sensitive CYP3A4 substrates and those with a narrow therapeutic range as defined by FDA are prohibited.

P-glycoprotein inhibitors

May increase serum concentration of temsirolimus and sirolimus which are P-glycoprotein substrates, or alter distribution. **Should be avoided or used with caution.**

P-glycoprotein inducers

May decrease serum concentration of temsirolimus and sirolimus which are P-glycoprotein substrates, or alter distribution. **Should be avoided or used with caution.**

ACE inhibitors

Risk of angioedema may be increased by temsirolimus. May be used with caution.

Cardiac glycosides

Temsirolimus may inhibit transport by P-glycoprotein. **Use is prohibited.**

Herbal/Nutriceutical

Avoid supplements containing St. John's Wort, alfalfa, aloe, bilberry, bitter melon, burdock, celery, damiana, echinacea, fenugreek, garcinia, garlic, ginseng (American), gymnema, marshmallow, stinging nettle.

Vaccines

Temsirolimus may diminish efficacy of vaccination and/or increase risk of toxicity from live vaccines. **Should be avoided** during protocol treatment.

6.7.2 Sorafenib

Sorafenib is metabolized by CYP3A4 and UGT1A9 and has been shown to inhibit CYP2B6, CYP2C8, and CYP2C9 as well as UGT1A1 and UGT1A9. Though inhibition of CYP2C9 has been documented, no effect was observed in an interaction study with warfarin. See Appendices 4-7.

CYP3A4 inhibitors

May increase serum concentration and toxicity of sorafenib. Strong inhibitors of CYP3A4 are prohibited; other CYP3A4 inhibitors should be avoided or used with caution.

CYP3A4 inducers

May decrease serum concentration of sorafenib. All inducers of CYP3A4 are prohibited.

Sensitive CYP3A, CYP2B6, CYP2C8, and CYP2C9 substrates and those with a narrow therapeutic range as defined by FDA are prohibited. (2009)

Other substrates of CYP3A4, CYP2B6, CYP2C8, and CYP2C9 should be avoided or used with caution.

UGT1A1 and UGT1A9 substrates

Sorafenib inhibits UGT1A1 and UGT1A9 and may increase exposure to these drugs; should be avoided or used with caution.

Doxorubicin

Sorafenib has been shown to increase levels of doxorubicin in vitro. Use is prohibited.

Herbal/Nutriceutical

Avoid echinacea and St. John's Wort as well as any herbal products which may interact with CYP3A4 system.

A detailed list of agents that should be **avoided or used with caution** due to risk for interaction with temsirolimus and sorafenib are in Appendices 6-9.

7. Reporting and Documentation of Results

7.1 Evaluation of Efficacy (or Activity)

7.1.1 Measurement of Response and Progression

Target and non-target lesions will be defined by RECIST guidelines (version 1.1): (Therasse, Arbuck et al. 2000) (Eisenhauer, Therasse et al.) All enrolled patients will imaging of chest, abdomen, and pelvis at baseline and after every 2 cycles (8 weeks ± 7 days) of protocol therapy. RECIST 1.1 measurements will be performed by centralized radiologist at UCSF, without knowledge of patients' clinical responses to protocol therapy, imaging studies during first 6 months on protocol therapy (including post-cycle 6 imaging), then at each study site by qualified investigator and/or radiologist for subsequent studies for patients who remain on protocol therapy. The schedule and methods of radiographic evaluation are described in previous sections.

7.1.1.1 Definitions

Evaluable for toxicity

All patients will be evaluable for toxicity from the time of their first treatment with the study drug. The study will use the CTCAE Version 4.0 for reporting adverse events.

Evaluable for efficacy

All enrolled patients who have received at least one dose of study drug (unless determined to be ineligible and replaced for reasons such as inability to comply or withdrawal of consent) will be evaluable for the primary efficacy endpoint according to the definitions stated below.

7.1.1.2 Disease Parameters

Measurable disease

Measurable disease is defined as lesions (or tumors) that can be accurately measured in at least one dimension (longest diameter to be recorded) with a minimum size of 10mm by CT scan (irrespective of scanner type) and MRI (no less than double the slice thickness and a minimum of 10mm), 10mm caliper measurement by clinical exam (when superficial), and/or 20mm by chest X-ray (if clearly defined and surrounded by aerated lung).

All tumor measurements will be recorded in millimeters or decimal fractions of centimeters. Previously irradiated lesions are considered non-measurable except in cases of documented progression of the lesion since the completion of radiation therapy.

Non-measurable disease

Non-measurable disease is all other lesions (or sites of disease), including small lesions (longest diameter < 20 mm with conventional techniques or < 10 mm using spiral CT scan). Leptomeningeal disease, ascites, pleural or pericardial effusion, inflammatory breast disease, lymphangitic involvement of skin or lung, abdominal masses/abdominal organomegaly identified by physical exam that is not measurable by reproducible imaging techniquesare all non-measurable.

Target lesions

All measurable lesions up to a maximum of 5 lesions total (and a maximum of two lesions per organ) representative of all involved organs should be identified as target lesions and recorded and measured at baseline. Target lesions will be selected on the basis of their size (lesions with the longest diameter) and their suitability for accurate repeated measurements (either by imaging techniques or clinically). A sum of the longest diameter (LD) for all target lesions will be calculated and reported as the baseline sum LD. The baseline sum LD will be used as reference by which to characterize the objective tumor response.

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. It is possible to record multiple non-target lesions involving the same organ as a single item on the case record form (e.g. “multiple enlarged pelvic lymph nodes” or “multiple liver metastases”). Measurements of these lesions are not required, but the presence or absence of each will be noted throughout follow-up.

7.1.1.3 Methods for Evaluation of Measurable Disease

All measurements will be taken and recorded in metric notation using a ruler or calipers. All baseline evaluations will be performed as closely as possible to the beginning of treatment and never more than 28 days before the beginning of the treatment.

The same method of assessment and the same technique will 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 when both methods have been used to assess the antitumor effect of a treatment.

Conventional CT and MRI

These techniques should be performed with cuts of 10 mm or less in slice thickness contiguously. Spiral CT should be performed using a 5 mm contiguous reconstruction algorithm. This applies to tumors of the chest, abdomen, and pelvis. Triphasic imaging of abdomen should be obtained when feasible.

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.

7.1.1.4 Response Criteria

Evaluation of Target Lesions

Complete Response (CR)

Disappearance of all target lesions, determined by two separate observations conducted not less than 4 weeks apart. Any pathological lymph nodes (whether target or non-target) must have reduction in short axis to <10 mm (the sum may not be “0” if there are target nodes). There can be no appearance of new lesions.

Partial Response (PR)

At least a 30% decrease in the sum of the longest diameter (LD) of target lesions, taking as reference the baseline sum LD. There can be no appearance of new lesions.

Progressive Disease (PD)

At least a 20% increase in the sum of the SLD of target lesions, taking as reference the smallest sum SLD recorded since the treatment started and minimum 5 mm increase over the nadir, or the appearance of one or more new lesions.

Stable Disease (SD)

Neither sufficient shrinkage to qualify for PR nor sufficient increase to qualify for PD, taking as reference the smallest sum LD since the treatment started.

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).

Incomplete Response/Stable Disease (SD)

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.

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.

Table 3.1.1.4 Response Criteria

Target Lesions	Non-Target Lesions	New Lesions	Overall Response	Best Response for this Category Also Requires
CR	CR	No	CR	> 4 weeks confirmation
CR	Non-CR/ Non-PD	No	PR	> 4 weeks confirmation
PR	Non-PD	No	PR	
SD	Non-PD	No	SD	documented at least once > 4 weeks from baseline
PD	Any	Yes or No	PD	no prior SD, PR or CR
Any	PD*	Yes or No	PD	
Any	Any	Yes	PD	

* In exceptional circumstances, unequivocal progression in non-target lesions may be accepted as disease progression

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 recurrent 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.

7.1.1.5 Centralized Review of Radiology

Radiographic assessments will be performed by a Radiologist at UCSF without knowledge of patients' clinical responses to protocol therapy for all patients' imaging studies during the first 6 months' on therapy (including imaging after completion of cycle 6). For patients enrolled on study at the Robert H. Lurie Comprehensive Cancer Center of Northwestern University, standard CT or MRI images will be acquired at that study site, stored on a CD as DICOM images, and shipped via FEDEX to the designated UCSF study coordinator. After cycle 6 imaging, RECIST 1.1 measurements may be performed at the treating site by a qualified investigator or radiologist. See Study Laboratory and Procedures Manual for detailed instructions.

Initial assessment of response or progression may be made based upon standard of care Radiology review plus clinical assessment if RECIST 1.1 measurements are not available at the time of study visit. In patients without signs of clinical progression, progression must be confirmed by RECIST 1.1 prior to removal from study. If progression is identified by RECIST 1.1 that was not identified by standard of care Radiology review plus clinical assessment, a patient will be removed from study at date of next scheduled study visit. Efforts will be made to obtain RECIST 1.1 measurements for patients treated at both sites within approximately 7 days of start of odd cycles.

7.1.2 Time to Event Endpoints

Time to Progression (TTP)

TTP is defined as the duration from date of first dose of protocol therapy to date of removal from study for radiographic (per RECIST 1.1) and/or clinical progression including death attributed to progression. For patients removed from study for other reasons than progression (such as toxicity, withdrawal of consent, death without documented progression, or other reasons without clinical or radiographic evidence of tumor progression), TTP will be censored at the date of study discontinuation.

Time to Treatment Failure (TTF)

TTF will be measured from date of first dose of protocol therapy to date of study discontinuation for progression, death, or toxicity. For patients removed from study for other reasons (such as noncompliance or withdrawal of consent), TTF will be censored at the date of study discontinuation.

Progression-Free Survival

Progression-free survival (PFS) is defined as the duration of time from date of first dose of protocol therapy to time of documented radiographic and/or clinical disease progression or death from any cause. Patients who have not progressed or died are censored at the date last known to be progression-free.

Overall Survival (OS)

Median OS for all enrolled patients who receive at least one dose of protocol therapy will be calculated from date of first dose of protocol therapy until date of death, using chart review and/or follow up phone calls to determine date of death in patients after removal from study. The survival of patients still alive after 5 years of follow up post study discontinuation will be censored. Alive patients are censored at the date last known alive.

7.2 Evaluation of Safety

Analyses will be performed for all patients having received at least one dose of study drug. The study will use the CTCAE Version 4.0 for reporting of non-hematologic adverse events and hematologic adverse events.

7.3 Evaluation of Exploratory Endpoints

Assessment of CTC enumeration, and patient blood specimen banking) is described below. Other exploratory endpoints (specifically, indices of viral hepatitis reactivation, and correlation between outcome measures and cause of viral hepatitis) will be measured/performed according to standard institutional procedures.

7.3.1 Circulating Tumor Cell Measurements

CTC will be enumerated using a novel slide-based assay (CEpiC) at baseline and on Day 1 of Cycles 2 and 3 (up to 7 days before treatment date). The slide-based circulating epithelial cell assay (CEpiC) is a new methodology to identify circulating epithelial cells and credential them as tumor cells. In the assay, each tube of approximately 7.5 mL (0.5 TBS) of anti-coagulated peripheral blood is lysed and all mononuclear cells are plated on cell adhesion glass slides. After appropriate pre-processing, the monolayer is incubated with a pan-anti-cytokeratin antibody and subsequently with a secondary fluorescent antibody. The samples are also stained with the nuclear stain DAPI. Slides are then analyzed with an automated digital microscope and data analyzed and correlated. The analysis predominantly consists of a cytometric and morphologic correlation that identifies intact cells that have a cytoplasmic cytokeratin stain consistent with epithelial cells. Controls include identically processed normal donor blood and sub-sampling of the patient blood excluding the primary antibody. The result of the CEpiC assay is the count of positive CEpiCs per milliliter of peripheral blood with associated imagery. CTC testing and analysis will be performed in the laboratory of Peter Kuhn, Ph.D., at the Scripps Research Institute, with the assistance of the clinical coordinator, Madelyn Luttgen. CTC testing will be performed without knowledge of the patients' clinical status.

Please see Study Laboratory and Procedures Manual for instructions regarding the collection, shipping, and handling of blood specimens for the CEpiC assay.

7.3.2 Markers of mTOR Pathway Activation

Archival tumor blocks will be accessioned then sectioned into 5 µm slides (3-5 plus H&E) and submitted to the UCSF Immunohistochemistry Core to perform IHC for selected mTOR pathway activation markers such as pAKT (S473), pS6RP (S235/236), PTEN (rabbit polyclonal antibody PN37), and LKB1 (D60C5). Optimization for each antibody will be performed using breast tumor samples as positive controls. The Director of the Core, [redacted], will read and provide reports for each case including intensity (0-3+), percentage positivity, distribution (very focal, focal, diffuse), and intracellular localization (membranous, cytoplasmic, nuclear) for each stain.[7-10]

In addition, FFPE sections from consenting patients will be submitted to the UCSF Clinical Cancer Genomics Lab (CCGL) for sequencing using hybridization-based target enrichment to analyze ~539 cancer-related genes for mutations (including mTOR pathway genes such as mTOR, AKT, TSC1/2, PTEN, PIK3CA) and copy number changes; selected genes will be analyzed for structural rearrangements (http://cancer.ucsf.edu/CCGL). Paired whole blood samples also will be sequenced for germline comparison.

7.3.3 Blood Specimen Banking

Blood samples totaling approximately 15-20 mL (1 to 1.3 TBS) optional patient blood samples (including serum, PBMC, whole blood, and plasma) will be obtained from all patients enrolled on this study additional potential future biomarker research. These blood samples will be taken at baseline, before temsirolimus dose on Cycle 1 Day 8, after completing 2 cycles of therapy (8 weeks ± 7 days), and at the end of treatment (if ≥ 4 weeks since last specimen(s) banked). For sample processing, handling, and shipping see Study Laboratory and Procedures Manual. The analyses will be performed by an investigator-approved laboratory.

7.3.4 Tumor Specimen Banking

Fresh specimen or formalin-fixed paraffin-embedded diagnosis tumor tissue and/or 10 to 20 unstained slides plus H&E slide from a tumor tissue block as well as corresponding pathology reports from initial diagnosis and/or subsequent tumor sampling will be retrospectively obtained. Specimens will be requisitioned from Pathology Department for banking as soon as possible after start of study. See Specimen Banking Procedures Manual.

8. Reporting and Documentation of Adverse Events

8.1 Definitions of Adverse Events

8.1.1 Adverse Event

An adverse event (also known as an adverse experience) is defined as any untoward medical occurrence associated with the use of a drug in humans, whether or not considered drug related. More specifically, an adverse event (can be any unfavorable and unintended sign (e.g., an abnormal laboratory finding), symptom, or disease temporally associated with the use of a drug, without any judgment about causality. An adverse event can arise from any use of the drug (e.g., off-label use, use in combination with another drug) and from any route of administration, formulation, or dose, including an overdose.

8.1.2 Adverse reaction

An adverse reaction is defined as any adverse event caused by the use of a drug. Adverse reactions are a subset of all suspected adverse reactions for which there is reason to conclude that the drug caused the event.

8.1.2.1 Suspected

A suspected adverse reaction is defined as any adverse event for which there is a reasonable possibility that the drug caused the adverse event. For the purposes of IND safety reporting, “reasonable possibility” indicates that there is evidence to suggest a causal relationship between the drug and the adverse event. A suspected adverse reaction implies a lesser degree of certainty about causality than an adverse reaction.

8.1.2.2 Unexpected

An adverse event or suspected adverse reaction is considered *unexpected* if it is not listed in the investigator brochure or package insert(s), or is not listed at the specificity or severity that has been observed, or, if an investigator brochure is not required or available, is not consistent with the risk information described in the general investigational plan or elsewhere in the current application.

“Unexpected,” as used in this definition, also refers to adverse events or suspected adverse reactions that are mentioned in the investigator brochure as occurring with a class of drugs or as anticipated from the pharmacological properties of the drug, but are not specifically mentioned as occurring with the particular drug under investigation.

Adverse events that would be anticipated to occur as part of the disease process are considered *unexpected* for the purposes of reporting because they would not be listed in the investigator brochure. For example, a certain number of non-acute deaths in a cancer trial would be anticipated as an outcome of the underlying disease, but such deaths would generally not be listed as a suspected adverse reaction in the investigator brochure.

Some adverse events are listed in the Investigator Brochure as occurring with the same class of drugs, or as anticipated from the pharmacological properties of the drug, even though they have not been observed with the drug under investigation. Such events would be considered *unexpected* until they have been observed with the drug under investigation. For example, although angioedema is anticipated to occur in some patients exposed to drugs in the ACE inhibitor class and angioedema would be described in the investigator brochure as a class effect, the first case of angioedema observed with the drug under investigation should be considered *unexpected* for reporting purposes.

8.1.2.3 Serious

An adverse event or suspected adverse reaction is considered *serious* if, in the view of either the investigator or sponsor, without regard to causality, it results in any of the following outcomes:

- Death
- Life-threatening adverse event
- Inpatient hospitalization or prolongation of existing hospitalization
- A persistent or significant disability or incapacity
- Congenital anomaly/birth defect
- Results in cancer

Important medical events that may not result in death, are life-threatening, or require hospitalization may be considered serious when, based upon appropriate medical judgment, they may jeopardize the patient or subject and may require medical or surgical intervention to prevent one of the outcomes listed in this definition. Examples of such medical events include allergic bronchospasm requiring intensive treatment in an emergency room or at home, blood dyscrasias or convulsions that do not result in inpatient hospitalization, or the development of drug dependency or drug abuse.

8.1.2.4 Life-threatening

An adverse event or suspected adverse reaction is considered *life-threatening* if, in the view of the sponsor-investigator, its occurrence places the patient or subject at immediate risk of death. It does not include an adverse event or suspected adverse reaction that, had it occurred in a more severe form, might have caused death.

8.2 Recording of an Adverse Event

All grade 3 and above adverse events will be entered into OnCore®, whether or not the event is believed to be associated with use of the study drug. Data about these events and their severity will be recorded using the NCI CTCAE v4.0.

The Investigator will assign attribution of the possible association of the event with use of the investigational drug, and this information will be entered into OnCore® using the classification system listed below:

Relationship	Attribution	Description
Unrelated to investigational drug/intervention	Unrelated	The AE *is clearly NOT related* to the intervention
	Unlikely	The AE *is doubtfully related* to the intervention
Related to investigational drug/intervention	Possible	The AE *may be related* to the intervention
	Probable	The AE *is likely related* to the intervention
	Definite	The AE *is clearly related* to the intervention

Signs or symptoms reported as adverse events will be graded and recorded by the Investigator according to the CTCAE. When specific adverse events are not listed in the CTCAE they will be graded by the Investigator as *none*, *mild*, *moderate* or *severe* according to the following grades and definitions:

Grade 0 No AE (or within normal limits)

Grade 1 Mild; asymptomatic or mild symptoms; clinical or diagnostic observations only; intervention not indicated

Grade 2 Moderate; minimal, local, or noninvasive intervention (e.g., packing, cautery) indicated; limiting age-appropriate instrumental activities of daily living (ADL)

Grade 3: Severe or medically significant but not immediately life-threatening; hospitalization or prolongation of hospitalization indicated; disabling; limiting self-care ADL

Grade 4: Life-threatening consequences; urgent intervention indicated

Grade 5: Death related to AE

8.3 Follow-up of Adverse Events

All adverse events will be followed with appropriate medical management until resolved. Patients removed from study for unacceptable adverse events will be followed until resolution or stabilization of the adverse event. For selected adverse events for which administration of the investigational drug was stopped, a re-challenge of the subject with the investigational drug may be conducted if considered both safe and ethical by the Investigator.

8.4 Adverse Events Monitoring

All adverse events, whether or not unexpected, and whether or not considered to be associated with the use of the study drug, will be entered into OnCore®, as noted above.

The Investigator will assess all adverse events and determine reportability requirements to the UCSF Data and Safety Monitoring Committee (DSMC) and UCSF's Institutional Review Board, the Committee on Human Research (CHR); and, when the study is conducted under an Investigational New Drug Application (IND), to the Food and Drug Administration (FDA) if it meets the FDA reporting criteria.

All adverse events entered into OnCore® will be reviewed by the Helen Diller Family Comprehensive Cancer Center Site Committee on a weekly basis. The Site Committee will review and discuss at each weekly meeting the selected toxicity, the toxicity grade, and the attribution of relationship of the adverse event to the administration of the study drug(s).

In addition, all adverse events and suspected adverse reactions considered "serious," entered into OnCore® will be reviewed and monitored by the Data and Safety Monitoring Committee on an ongoing basis, discussed at DSMC meetings which take place every six (6) weeks.

8.5 Expedited Reporting

Reporting to the Data and Safety Monitoring Committee

If a death occurs during the treatment phase of the study or within 30 days after the last administration of the study drug(s) and it is determined to be related either to the study drug(s) or to a study procedure, the Investigator or his/her designee must notify the DSMC Chair (or qualified alternate) within 1 business day of knowledge of the event. The contact may be by phone or e-mail.

Reporting to UCSF Committee on Human Research (Institutional Review Board)

The Principal Investigator must report events meeting the UCSF CHR definition of "Unanticipated Problem" (UP) within 5 business days of his/her awareness of the event.

Expedited Reporting to the Food and Drug Administration

If the study is being conducted under an IND, the Sponsor-Investigator is responsible for determining whether or not the suspected adverse reaction meets the criteria for expedited reporting in accordance with Federal Regulations (21 CFR §312.32).

The Investigator must report in an IND safety report any suspected adverse reaction that is both serious and unexpected. The Sponsor-Investigator needs to ensure that the event meets all three definitions:

- Suspected adverse reaction (as defined in 8.1.2.1)
- Unexpected (as defined in 8.1.2.2)
- Serious (as defined in 8.1.2.3)

If the adverse event does not meet all three of the definitions, it should not be submitted as an expedited IND safety report.

The timeline for submitting an IND safety report to FDA is no later than **15 calendar days** after the Investigator determines that the suspected adverse reaction qualifies for reporting (21 CFR 312.32(c)(1)).

Any unexpected fatal or life-threatening suspected adverse reaction will be reported to FDA no later than **7 calendar days** after the Investigator's initial receipt of the information (21 CFR 312.32(c)(2)).

Any relevant additional information that pertains to a previously submitted IND safety report will be submitted to FDA as a Follow-up IND Safety Report without delay, as soon as the information is available (21 CFR 312.32(d)(2)).

8.6 Adverse Event Reporting to Funding Organization and Research Grant Provider

In addition to complying with all applicable regulatory reporting laws and regulations, each site will report the following information in writing to NCCN and research grant provider (Pfizer) in writing within 24 hours of the Investigator's awareness of occurrence. Principal Investigator should report SAEs as soon as they are determined to meet the definition, even if complete information is not yet available:

a) a death, regardless of whether it is considered related to treatment with the Study Drug

b) a non-fatal SAE that occurs during the reporting period and that is assessed by the Principal Investigator as both related to treatment with the Study Drug and unexpected for that product

c) an SAE assessed by the Principal Investigator as related to the Study Drug that occurs after the SAE reporting period,

d) an otherwise reportable event - Exposure During Pregnancy, Exposure During Lactation, Occupational Exposure, and Lack Of Effect. Even though there may not be an associated SAE, exposure to the Study Drug during pregnancy, exposure to the Study Drug during lactation, and occupational exposure to the Study Drug are reportable, and lack of effect of the Study Drug may also be reportable.

An event should be considered "related" to the Study Drug if a relationship is at least a reasonable possibility, and "unexpectedness" should be based upon a single safety reference document identified by the Principal Investigator and documented in association with the study.

Follow-up information regarding any of the above

- The participating investigator should include his or her assessment of the causal relationship between each SAE and the research grant provider's product.
- Reports to NCCN will include the cover page provided (see Appendix J attached hereto), and reference Protocol Number NCCN-T12. Submit to NCCN via fax at 1-215-358-7699 or via e-mail to ORPReports@nccn.org
- Reports to Pfizer, Inc. will include the Reportable Event Fax Cover sheet (see Appendix 3.1) (use NCCN-T12 as the external reference number). Submit to the Pfizer U.S. Clinical Trial Department [redacted].
- A copy of the report should be sent to the coordinating center CRC

9. Statistical Considerations and Evaluation of Results

9.1 Study Endpoints

9.1.1 Study Design

This study is a single-arm, one-stage, open-label phase II trial to evaluate the efficacy of the combination of temsirolimus plus sorafenib in first line therapy for patients with histologically-confirmed advanced HCC.

9.1.2 Primary Endpoint

Median TTP will be calculated in months from date of first dose of protocol therapy to date of removal from study for radiographic (per RECIST 1.1) and/or clinical progression including death attributed to progression. For patients removed from study for other reasons than progression (such as toxicity, withdrawal of consent, or death from other causes than documented radiographic and/or clinical progression), TTP will be censored at the date last known to be progression-free.

9.1.3 Secondary Endpoints

1. Response rate (RR) will be measured by RECIST version 1.1.

2. Median PFS will be calculated in months from date of first dose of protocol therapy to date of study removal for documented radiographic and/or clinical disease progression or death from any cause. For patients removed from study for other reasons than progression or death, PFS will be censored at the date last known to be progression-free.

3. Median OS for all enrolled patients who receive at least one dose of protocol therapy will be calculated from date of first dose of protocol therapy until date of death, using chart review and/or follow up phone calls to determine date of death in patients after removal from study. The survival of patients still alive after 5 years of follow up post study discontinuation will be censored.

4. TTF will be measured from date of first dose of protocol therapy to date of removal from study for progression, death, or toxicity. For patients removed from study for other reasons (such as noncompliance or withdrawal of consent), TTF will be censored at the date of study discontinuation.

5. In patients with baseline AFP ≥ 20 ng/mL, AFP response will be measured by the percent change from baseline value to the value at the time of best AFP response. The proportion of ≥ 50% decline from baseline will be measured. The change in AFP will also be examined for association with absolute TTP and best response.

6. Toxicity will be measured using Common Terminology Criteria for Adverse Events CTCAE) version 4.0. The frequency of dose reduction, treatment delays, and discontinuation for lack of tolerability will be quantified.

9.1.4 Exploratory Endpoints

1. TTP, RR, PFS, OS, TTF, AFP response, and toxicity and tolerability will be described in subsets defined by HBV, HCV, Asian, and non-Asian status.

2. CTC will be enumerated in peripheral blood at baseline and after 1 and 2 cycles of therapy to evaluate for any relationship between baseline levels and change in levels with median TTP and/or OS.

3. Archival tumor specimens will be sectioned for IHC testing of selected mTOR pathway markers and next-generation sequencing. Markers will be dichotomized as high or low using published cutpoints to describe incidence of activation in study population and explore for relationship to clinical outcomes including TTP and AFP response.

4. In patients with chronic active HBV and/or positive HBcAb, hepatitis B viral DNA quantitative will be monitored for rates of viremia/reactivation on study.

5. Optional patient blood specimens (including serum, PBMC, whole blood, and plasma) and left-over archival pathology specimens will be banked in the UCSF Hepatobiliary Tissue Bank (HBTB) for future research.

9.2 Determination of Sample Size and Accrual Rate

9.2.1 Sample Size and Power Estimate

The sample size for this phase II study will be 25 patients under the assumption that the time to progression is approximately exponentially distributed and patients come onto study uniformly

during the study accrual. A sample size of 25 evaluable patients will have sufficient power to detect the difference between the null hypothesis that the median TTP is less than 3 months (based on historical controls) and the alternative hypothesis that the median TTP is more than 6 months. The study will have a 1-sided significance level of 10% and a power of 88% under the exact test for comparison of the TTP rates between p0 < 25% versus p1 > 50% at 6 months.

A median TTP of > 6 months would constitute a meaningful outcome in advanced HCC, based upon the benchmarks of first-line TTP of 5.5 months in the SHARP study and 2.8 months in the Asia-Pacific study.(Llovet, Ricci et al. 2008; Cheng, Kang et al. 2009) If median TTP is > 6 months in this study, a randomized phase II trial to compare the combination therapy to single-agent sorafenib should be considered. Such a trial would be much larger and more costly, however, and should not be undertaken prior to the current study to confirm that there is tolerability and a signal of efficacy to warrant proceeding with the larger trial, noting in particular the challenges to accrual in first-line HCC studies historically, as faced by contemporary cooperative group studies CALGB 80802 and ECOG 1208.

Accrual of 25 paired archival tumor and blood specimens also constitutes a meaningful endpoint in that it will enable future biomarker studies in subsets of patients with radiographic response, AFP response, prolonged stable disease, and primary progression, including potential exploratory studies for association with tissue biomarkers of mTOR pathway activation (such as pS6 staining).(Villanueva, Chiang et al. 2008)

9.2.2 Accrual estimates

It is estimated that 12 patients will be accrued annually (approximately 1 patient per month) combined between UCSF and Northwestern. This estimate is based upon the accrual to the predecessor Phase I study of this combination which enrolled 25 patients with similar eligibility criteria between December 2009 and April 2012. It is expected that the Phase II study will accrue faster than the Phase I due to the established infrastructure and momentum from the Phase I study, along with no requirement for delays for dose escalation and frequent amendments which were associated with the Phase I study.

9.3 Interim Analyses and Stopping Rules

No interim assessment for primary endpoint is planned, but there will be a planned interim safety analysis by the UCSF Data and Safety Monitoring Board after the first 30% of the planned study participants (8 patients) have enrolled and completed at least one cycle of protocol therapy or been removed for toxicity. Study accrual will not be interrupted for the early safety assessment. The proportion of patients with treatment-related adverse events of grade 3 or higher and/or an unacceptable toxicity event (with exceptions defined in Section 6.4) will be summarized with 90% two-sided exact confidence intervals. Each confidence interval will be no wider than ±0.17. If the lower confidence bound (LCB) exceeds 0.17, adverse events will be evaluated and a decision will be made whether to amend the protocol or close the study.

If the true rate of treatment-related toxicity is 25%, the probability that 3 or more patients will have adverse events of grade 3 or greater is 32%. A summary of probabilities for a range of true toxicity rates is shown below. If 3 or more patients among the first 8 experience an eligible grade 3 or higher and/or unacceptable toxicity event, a decision will be made whether to amend the study or close the study due to toxicity.

True Toxicity Rate	***Probability 3 or more of 8 patients experience AEs of grade 3 or greater***	***Probability 2 or more of 8 patients experience AEs of grade 3 or greater***	***Probability 1 or more of 8 patients experience AEs of grade 3 or greater***

10%	3.8%	18.7%	57%
20%	20.3%	50.0%	83%
25%	32.0%	63.3%	89%
30%	44.8%	74.5%	94%

9.4 Analyses Plans

Analyses of primary and secondary endpoints as well as exploratory analyses will be conducted by [redacted] the designated biostatistician for this protocol. Demographic and other baseline characteristics will be summarized in the aggregate manner. For continuous demographic/baseline variables including age, weight, and vital signs, results will be summarized and presented as N, mean, standard deviation, median, and minimum and maximum values. For categorical variables such as race/ethnicity, the number and percentage of subjects will be used. Statistical comparisons of these baseline characteristics will be performed using the Wilcoxon rank-sum test for continuous variables and Fisher's exact test for categorical variables.

9.4.1 Analysis Population

The safety population will include all enrolled patients who receive at least one dose of the study drug. All patients will be evaluable for toxicity and safety assessment from the time of their first treatment with the study drug.

The evaluable population for efficacy will include all enrolled patients who have received at least one dose of study drug (unless determined to be ineligible and replaced for reasons such as inability to comply or withdrawal of consent). These patients will have their response classified according to the definitions stated below.

9.4.2 Analysis of Primary Endpoints

Kaplan-Meier methods will be used to summarize the primary endpoint (median TTP) with 95% confidence intervals. The proportion of patients with TTP equal to or exceeding 6 months will also be calculated and reported along with 95% confidence intervals.

9.4.3 Analysis of Secondary Endpoints

Response rates will be summarized and presented with 95% exact binomial confidence intervals. The comparison of response rates will use Fisher's exact test.

Kaplan-Meier methods will be used to summarize time-to-event outcomes including the primary endpoint (median TTP) and secondary endpoints (PFS, TTF, OS). Proportions of patients with TTP, PFS, and TTF equal to or exceeding 6 months will be calculated with 95% confidence intervals. Tumor response by RECIST 1.1 measurements will be presented descriptively for the study cohort.

- AFP Response: Baseline levels (high or low) will be compared to absolute TTP to determine if there is an association using Fisher's Exact tests or by Wilcoxon Rank-Sum test. Changes in levels on therapy with temsirolimus and sorafenib will be measured as an increase by ≥ 50% from baseline, stable value within 50% of baseline, or decrease by ≥ 50% from baseline. The change in level will be used to evaluate the predictive value for TTP. Fisher's Exact tests will be used to test for significance. Kaplan-Meier methods and Log-rank tests will also be used to estimate TTP based on the AFP.

9.4.4 Exploratory Analyses/Assessments

- CTC levels: Baseline level of CTC (detectable or undetectable) will be compared to proportion with TTP at 6 months to determine if there is an association using Fisher's Exact tests. Change in CTC levels (increase or decrease) after 1 and 2 cycles will be compared to proportion with TTP at 6 months to determine if there is an association using Fisher's Exact tests. Markers of mTOR pathway activation: Descriptive statistics will summarize the IHC parameters of staining intensity, percentage positivity, distribution, and intracellular localization for each protein. Due to possible limited specimen quantity, IHC outcomes will be dichotomized. Activation will be measured by % positive cells and intensity using published IHC cutpoints for high/low: cytoplasmic pAkt > 40% or 2-3+, cytoplasmic PTEN intensity < 10% or 0-1+, cytoplasmic pS6RP > 70% or 2-3+, or LKB1 0-1.[7-12] Univariate analysis using Cox's model will be performed to test whether the dichotomized expression of each protein using the published cutpoints is a significant predictor of TTP. Results for each test will include the hazard ratio (HR) with 95% confidence intervals. The association between each protein and the two binary coding schemes for AFP decline (≥ 50% or ≥ 20%) on C2D1 will be tested using Fisher's exact test. Results for each test will be summarized with proportions with pathway activation and 95% confidence intervals for each AFP subset. Each patient will also be categorized as having overall mTOR pathway activation (defined as any combination of high pAKT, high pS6RP, low PTEN, and/or low LKB1) or not. This binary variable will also be examined for its relationship with TTP, OS, AFP response, and other clinical endpoints using the same methods described above. For each test of association with AFP change, based upon Fisher's exact test large differences can be detected (e.g. 10% among 7 patients with marker-low, 80% among 8 patients with marker-low) (power=81%, α_2=10%). The sample size is too small to test for a difference in TTP, OS, or other clinical endpoints according to pathway activation status. The expected sample size will be ≥ 25 cases (expect 40 to 45 cases) with IHC-quality archival tumor, with the expectation that ~75% of cases will be evaluable for TTP (n ≥ 18) and 60% evaluable for AFP response on C2D1 (n ≥ 15).

  Genotype findings will be dichotomized as presence or absence of any mTOR pathway aberration or combination thereof (e.g. mutations in AKT, mTOR, PTEN, PIK3CA, and/or TSC1/2). This binary variable will be examined for its relationship with time to progression, overall survival, AFP response, and other clinical endpoints by univariate analysis using Cox's model. Concordance between primary and metastatic specimens on mutational profile will be described individually due to small sample size. Other tumor genotype findings and relationship to clinicopathologic covariates will be reported descriptively.

  To optimize sample size and ensure adequate number of evaluable cases, these analyses of mTOR pathway IHC and genotype markers will also be performed on cases and samples CC#09455 when available, to ensure at least 25 evaluable cases with adequate archival tumor for testing. Although pooling of data and specimens across studies introduces potential confounding, it is appropriate for these exploratory analyses as the eligibility criteria and protocol therapy were overlapping for CC#09455 and the current study.
- Hepatitis B viral load: In all patients with active HBV infection, changes in HBV DNA quantitative by PCR (conversion from undetectable at baseline to detectable on study or an increase by ≥ 50% from baseline versus unchanged from baseline) will be characterized using descriptive statistics (mean, median, standard deviation, and 95% confidence interval) to calculate the proportion of patients with an increase in viral load while on protocol therapy.
- Future biomarker research on banked specimens will be reported outside of the main study report

9.5 Evaluation of Safety

Analyses will be performed for all patients having received at least one dose of study drug. The study will use the NCI Common Terminology Criteria for Adverse Events (CTCAE) Version 4.0.

10. Study Management

10.1 Pre-study Documentation

This study will be conducted in accordance with the ethical principles that have their origin in the Declaration of Helsinki as stated in 21 CFR §312.120(c)(4); consistent with GCP and all applicable regulatory requirements.

Before initiating this trial, the Investigator will have written and dated approval from the Institutional Review Board for the protocol, written informed consent form, subject recruitment materials, and any other written information to be provided to subjects before any protocol related procedures are performed on any subjects.

The clinical investigation will not begin until either FDA has determined that the study under the Investigational Drug Application (IND) is allowed to proceed or the Investigator has received a letter from FDA stating that the study is exempt from IND requirements.

The Investigator must comply with the applicable regulations in Title 21 of the Code of Federal Regulations (21 CFR §50, §54, and §312), GCP/ICH guidelines, and all applicable regulatory requirements. The IRB must comply with the regulations in 21 CFR §56 and applicable regulatory requirements.

10.2 Institutional Review Board Approval

The protocol, the proposed informed consent form, and all forms of participant information related to the study (e.g. advertisements used to recruit participants) will be reviewed and approved by the UCSF CHR (UCSF Institutional Review Board). Prior to obtaining CHR approval, the protocol must be approved by the Helen Diller Family Comprehensive Cancer Center Site Committee and by the Protocol Review Committee (PRC). The initial protocol and all protocol amendments must be approved by the IRB prior to implementation.

10.3 Informed Consent

All participants must be provided a consent form describing the study with sufficient information for each participant to make an informed decision regarding their participation. Participants must sign the CHR-approved informed consent form prior to participation in any study specific procedure. The participant must receive a copy of the signed and dated consent document. The original signed copy of the consent document must be retained in the medical record or research file.

10.4 Changes in the Protocol

Once the protocol has been approved by the UCSF CHR, any changes to the protocol must be documented in the form of an amendment. The amendment must be signed by the Investigator and approved by PRC and the CHR prior to implementation.

If it becomes necessary to alter the protocol to eliminate an immediate hazard to patients, an amendment may be implemented prior to CHR approval. In this circumstance, however, the Investigator must then notify the CHR in writing within five (5) working days after implementation. The Study Chair and the UCSF study team will be responsible for updating any participating sites.

10.5 Handling and Documentation of Clinical Supplies

The UCSF Principal Investigator and each participating site will maintain complete records showing the receipt, dispensation, return, or other disposition of all investigational drugs. The date, quantity and batch or code number of the drug, and the identification of patients to whom study drug has been dispensed by patient number and initials will be included. The sponsor-investigator will maintain written records of any disposition of the study drug.

The Principal Investigator shall not make the investigational drug available to any individuals other than to qualified study patients. Furthermore, the Principal Investigator will not allow the investigational drug to be used in any manner other than that specified in this protocol.

10.6 Case Report Forms (CRFs)

The Principal Investigator and/or his/her designee, will prepare and maintain adequate and accurate participant case histories with observations and data pertinent to the study. Study specific Case Report Forms (CRFs) will document safety and treatment outcomes for safety monitoring and data analysis. All study data will be entered into OnCore® via standardized CRFs in accordance with the CTMS study calendar, using single data entry with a secure access account. The Clinical Research Coordinator (CRC) will complete the CRFs as soon as possible upon completion of the study visit; the Investigator will review and approve the completed CRFs.

The information collected on CRFs shall be identical to that appearing in original source documents. Source documents will be found in the patient's medical records maintained by UCSF personnel. All source documentation should be kept in separate research folders for each patient.

In accordance with federal regulations, the Investigator is responsible for the accuracy and authenticity of all clinical and laboratory data entered onto CRFs. The PI will approve all completed CRFs to attest that the information contained on the CRFs is true and accurate.

All source documentation and CTMS data will be available for review/monitoring by the UCSF DSMC and regulatory agencies.

The Principal Investigator will be responsible for ensuring the accurate capture of study data. At study completion, when the CRFs have been declared to be complete and accurate, the database will be locked. Any changes to the data entered into the CRFs after that time can only be made by joint written agreement among the Study Chair, the Trial Statistician, and the Protocol Project Manager.

10.7 Oversight and Monitoring Plan

The UCSF Helen Diller Family Comprehensive Cancer Center DSMC will be the monitoring entity for this study. The UCSF DSMC will monitor the study in accordance with the NCI-approved Data and Safety Monitoring Plan (DSMP). The DSMC will routinely review all adverse events and suspected adverse reactions considered "serious". The DSMC will audit study-related activities to ensure that the study is conducted in accordance with the protocol, local standard operating procedures, FDA regulations, and Good Clinical Practice (GCP). Significant results of the DSMC audit will be communicated to the IRB and the appropriate regulatory authorities at the time of continuing review, or in an expedited fashion, as applicable. See Appendix 3, Data and Safety Monitoring Plan for additional information.

10.8 Multicenter communication

The UCSF Coordinating Center provides administration, data management, and organizational support for the participating sites in the conduct of a multicenter clinical trial. The UCSF Coordinating Center will also coordinate, at minimum, monthly conference calls with the participating sites at the completion of each cohort or more frequently as needed to discuss risk assessment. The following issues will be discussed as appropriate:

- Enrollment information
- Cohort updates (i.e. DLTs)
- Adverse events (i.e. new adverse events and updates on unresolved adverse events and new safety information)
- Protocol violations
- Other issues affecting the conduct of the study

10.9 Record Keeping and Record Retention

The Principal Investigator is required to maintain adequate records of the disposition of the drug, including dates, quantity, and use by subjects, as well as written records of the disposition of the drug when the study ends.

The Principal Investigator is required to prepare and maintain adequate and accurate case histories that record all observations and other data pertinent to the investigation on each individual administered the investigational drug or employed as a control in the investigation. Case histories include the case report forms and supporting data including, for example, signed and dated consent forms and medical records including, for example, progress notes of the physician, the individual's hospital chart(s), and the nurses' notes. The case history for each individual shall document that informed consent was obtained prior to participation in the study.

Study documentation includes all CRFs, data correction forms or queries, source documents, Sponsor-Investigator correspondence, monitoring logs/letters, and regulatory documents (e.g., protocol and amendments, CHR correspondence and approval, signed patient consent forms).

Source documents include all recordings of observations or notations of clinical activities and all reports and records necessary for the evaluation and reconstruction of the clinical research study.

In accordance with FDA regulations, the investigator shall retain records for a period of 2 years following the date a marketing application is approved for the drug for the indication for which it is being investigated; or, if no application is to be filed or if the application is not approved for such indication, until 2 years after the investigation is discontinued and FDA is notified.

10.10 Coordinating Center Documentation of Distribution

It is the responsibility of the Study Chair to maintain adequate files documenting the distribution of study documents as well as their receipt (when possible). The HDFCCC recommends that the Study Chair maintain a correspondence file and log for each segment of distribution (e.g., FDA, drug manufacturer, participating sites, etc.).

Correspondence file: should contain copies (paper or electronic) of all protocol versions, cover letters, amendment outlines (summary of changes), etc., along with distribution documentation and (when available) documentation of receipt.

Correspondence log: should be a brief list of all documents distributed including the date sent, recipient(s), and (if available) a tracking number and date received.

At a minimum, the Study Chair must keep documentation of when and to whom the protocol, its updates and safety information are distributed.

10.11 Regulatory Documentation

Prior to implementing this protocol at UCSF HDFCCC, the protocol, informed consent form, HIPAA authorization and any other information pertaining to participants must be approved by the UCSF Committee on Human Research (CHR). Prior to implementing this protocol at the participating sites, approval for the UCSF CHR approved protocol must be obtained from the participating site's IRB.

The following documents must be provided to UCSF HDFCCC before the participating site can be initiated and begin enrolling participants:

- Participating Site IRB approval(s) for the protocol, appendices, informed consent form and HIPAA authorization
- Participating Site IRB approved consent form
- Participating Site IRB membership list
- Participating Site IRB's Federal Wide Assurance number and OHRP Registration number
- Curriculum vitae and medical license for each investigator and consenting professional
- Documentation of Human Subject Research Certification training for investigators and key staff members at the Participating Site
- Participating site laboratory certifications and normals

Upon receipt of the required documents, UCSF HDFCCC will formally contact the site and grant permission to proceed with enrollment.

11. References

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Appendix 1 ECOG Performance Status Criteria

ECOG Performance Status Scale	
Grade	*Descriptions*
0	Normal activity Fully active, able to carry on all pre-disease performance without restriction
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)
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
3	In bed > 50% of the time Capable of only limited self-care, confined to bed or chair more than 50% of waking hours
4	100% bedridden Completely disabled Cannot carry on any self-care Totally confined to bed or chair
5	Dead

Appendix 2 Staging Tools

Child-Pugh Score

http://homepage.mac.com/sholland/contrivances/childpugh.html

Below is an example of a calculated score:

Naperville Gastroenterology

Stephen Holland, MD, FACP
636 Raymond Drive, Suite 201
Naperville, IL 60563

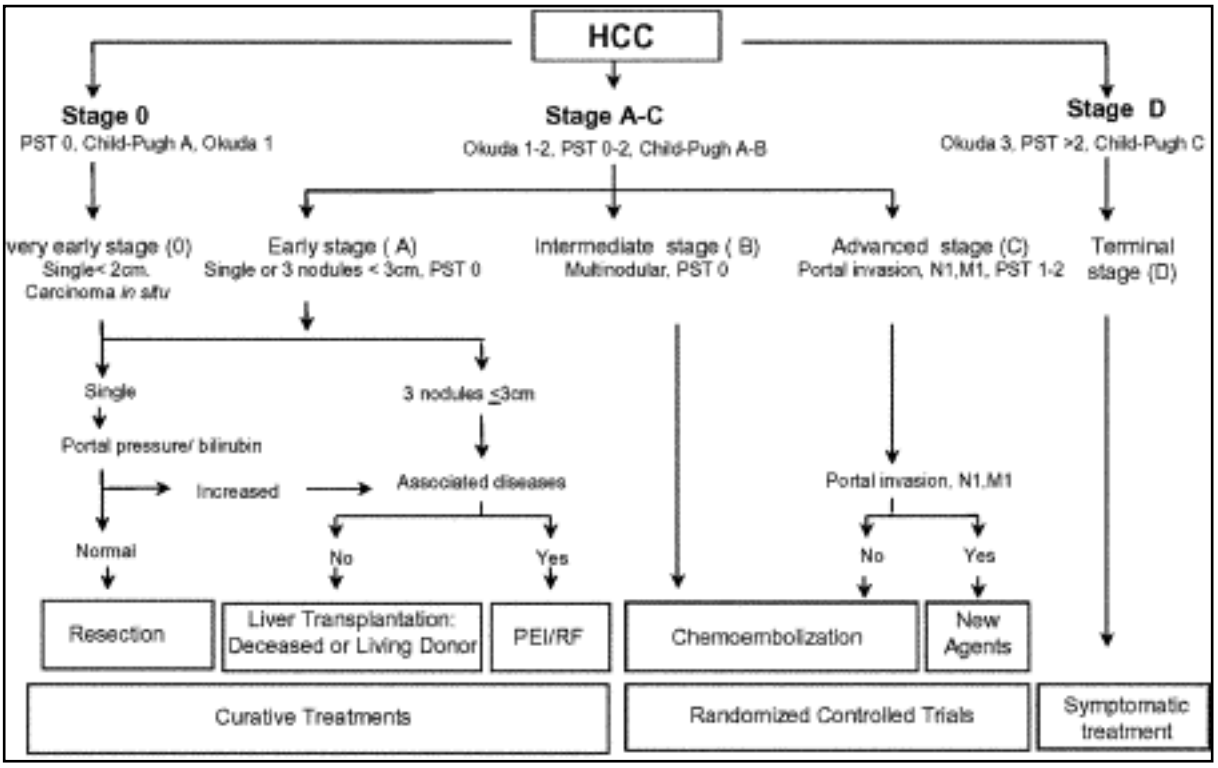
Child-Pugh Score Calculator

Bilirubin: ⦿<2 mg/dl (34 umol/l) ○2-3 mg/dl (34-50 umol/l) ○>3 mg/dl (50 umol/l)
Albumin: ⦿>3.5 g/dl ○3.5-2.8 ○<2.8
PT prolongation (INR): ⦿<4 seconds (<1.7)
○4-6 seconds (1.7-2.3)
○>6 seconds (>2.3)
Ascites: ⦿Absent ○Mild-Moderate ○Severe/Refractory
Encephalopathy: ⦿Absent ○Mild (I-II) ○Severe (III-IV)

Child-Pugh Score: 5

Interpretation:
Class A: 5-6
Class B: 7-9
Class C: 10-15

Barcelona Clinic Liver Cancer (BCLC) Stage



Barcelona-Clinic Liver Cancer (BCLC) staging classification and treatment schedule

Stage 0: Patients with very early HCC are optimal candidates for resection. Stage A: Patients with early HCC are candidates for radical therapies (resection and ablation, liver transplantation; or percutaneous treatments). Stage B: Patients with intermediate HCC may benefit from chemoembolization. Stage C: Patients with advanced HCC may receive new agents in the setting of a RCT. Stage D: Patients with end-stage disease will receive symptomatic treatment. (Adapted from Llovet JM et al with permission.)

Liver Transplantation Volume 10, Issue S2, pages S115-S120, 30 JAN 2004 DOI: 10.1002/lt.20034
http://onlinelibrary.wiley.com/doi/10.1002/lt.20034/full#fig1

Cancer of the Liver Program (CLIP) Score

TABLE 1. CLIP Scoring System

Variable	Score
Child-Pugh stage	
A	0
B	1
C	2
Tumor morphology	
Uninodular and extension ≤50%	0
Multinodular and extension ≤50%	1
Massive or extension >50%	2
AFP	
<400	0
≥400	1
Portal vein thrombosis	
No	0
Yes	1

"Prospective Validation of the CLIP Score: A New Prognostic System for Patients With Cirrhosis and Hepatocellular Carcinoma".The Cancer of The Liver Italian Program (CLIP) Investigators Hepatology Vol. 31, No. 4, 2000
http://onlinelibrary.wiley.com/doi/10.1053/he.2000.5628/pdf

Appendix 3 Data and Safety Monitoring Plan for Multicenter Institutional Study (Phase 2 or 3 Institutional Study)

The UCSF Helen Diller Family Comprehensive Cancer Center (HDFCCC) Data and Safety Monitoring Committee (DSMC) is responsible for monitoring data quality and subject safety for all HDFCCC institutional clinical studies. A summary of DSMC activities for this study includes:

- Review of subject data
- Review of suspected adverse reactions considered "serious"
- Monthly monitoring (depending on study accrual)
- Minimum of a yearly regulatory audit

Monitoring and Reporting Guidelines

All institutional Phase 2 or 3 therapeutic studies are designated with a moderate risk assessment. The data is monitored every six months, with twenty percent of the subjects monitored (or at least three subjects if the calculated value is less than three).

The UCSF Coordinating Center provides administration, data management, and organizational support for the participating sites in the conduct of a multicenter clinical trial. The UCSF Coordinating Center will also coordinate quarterly conference calls with the participating sites to communicate the review of adverse events, safety data, and other study matters.

The Principal Investigator at the UCSF Coordinating Center will hold the role of Study Chair. The Study Chair is responsible for the overall conduct of the study and for monitoring its safety and progress at all participating sites. The Study Chair will conduct continuous review of data and subject safety and discuss each subject's treatment at monthly UCSF Site Committee meetings. The discussions are documented in the UCSF Site Committee meeting minutes.

<u>Multicenter communication</u>

The UCSF Coordinating Center provides administration, data management, and organizational support for the participating sites in the conduct of a multicenter clinical trial. The UCSF Coordinating Center will also coordinate, at minimum, monthly conference calls with the participating sites at the completion of each cohort or more frequently as needed to discuss risk assessment. The following issues will be discussed as appropriate:

- Enrollment information
- Adverse events (i.e. new adverse events and updates on unresolved adverse events and new safety information)
- Protocol violations
- Other issues affecting the conduct of the study

Adverse events reporting to the DSMC will include reports from both the UCSF Coordinating Center, as well as the participating sites. The DSMC will be responsible for monitoring all data entered in OnCore® at the UCSF Coordinating Center and the participating sites. The data (i.e. copies of source documents) from the participating sites will be sent electronically or faxed over to the UCSF Coordinating Center prior to the monitoring visits in order for the DSMC to monitor the participating site's compliance with the protocol, patient safety, and to verify data entry.

Adverse Event Review and Monitoring

Adverse Event Monitoring

All Grade 3-5 Adverse Events, whether or not unexpected, and whether or not considered to be associated with the use of study drug, will be entered into OnCore®, UCSF's Clinical Trial Management System.

All Adverse Events will be reported by the participating site to the Coordinating Center (UCSF) approximately weekly, as required for weekly review of study data and as per local IRB guidelines within the required timeframes.

All Grade 3-5 adverse events entered into OnCore® will be reviewed on a monthly basis at the UCSF Site Committee meetings. All clinically significant adverse events must be reported to the UCSF Coordinating Center by the participating sites within 10 business days of becoming aware of the event or during the next scheduled quarterly conference call, whichever is sooner. The UCSF Site Committee will review and discuss the selected toxicity, the toxicity grade, and the attribution of relationship of the adverse event to the administration of the study drug(s) from the UCSF Coordinating Center and the participating sites.

In addition, all suspected adverse reactions considered "serious" must be entered in OnCore® and reported to the UCSF Coordinating Center within 1 business day. The suspected adverse reactions considered "serious" will be reviewed and monitored by the Data and Safety Monitoring Committee on an ongoing basis and discussed at the DSMC meeting, which take place every six (6) weeks.

If a death occurs during the treatment phase of the study or within 30 days after the last administration of the study drug(s) and is determined to be related either to the investigational drug or any research related procedure, the Study Chair at the UCSF Coordinating Center or the assigned designee must be notified within 1 business day from the participating site(s) and the Study Chair must then notify the DSMC Chair or qualified alternate within 1 business day of this notification. The contact may be by phone or e-mail.

Increase in Adverse Event Rates

If an increase in the frequency of Grade 3 or 4 adverse events (above the rate reported in the Investigator Brochure or package insert), the Study Chair at the UCSF Coordinating Center is responsible for notifying the DSMC at the time the increased rate is identified. The report will indicate if the incidence of adverse events observed in the study is above the range stated in the Investigator Brochure or package insert.

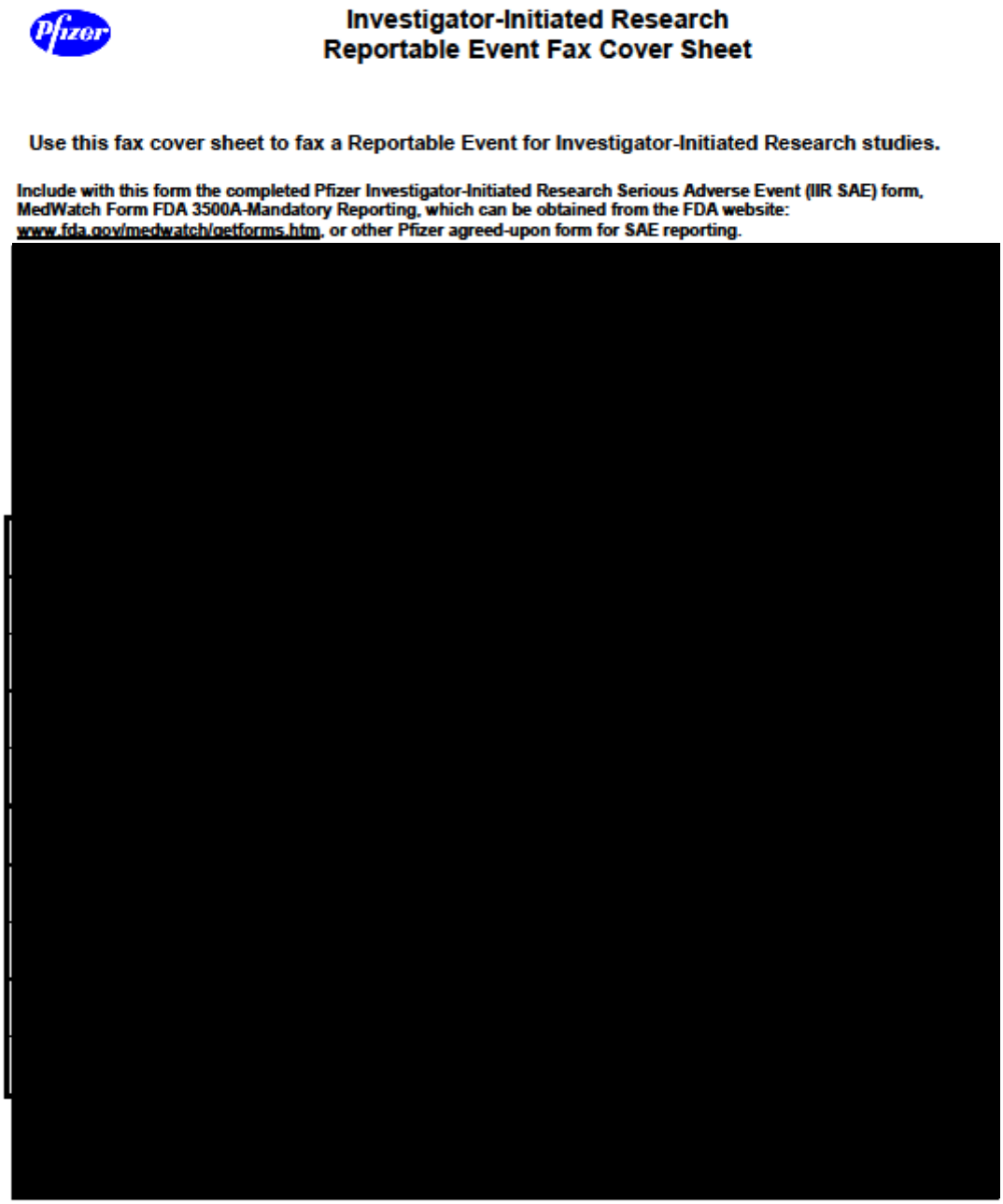
If at any time the Study Chair stops enrollment or stops the study due to safety issues, the DSMC Chair and DSMC Manager must be notified within 1 business day via e-mail. The DSMC must receive a formal letter within 10 business days and the CHR must be notified.

Data and Safety Monitoring Committee Contacts:

DSMC Chair: [redacted]
Phone: [redacted]
Email: [redacted]
Address: Box 1705
UCSF
San Francisco, CA 94115

DSMC Monitors
Box 1297
UCSF Helen Diller Family Comprehensive Cancer Center
San Francisco, CA 94115

3.1 Cover Sheet for SAE Reporting to NCCN and Research Grant Provider



Investigator-Initiated Research
Reportable Event Fax Cover Sheet

Use this fax cover sheet to fax a Reportable Event for Investigator-Initiated Research studies.

Include with this form the completed Pfizer Investigator-Initiated Research Serious Adverse Event (IIR SAE) form, MedWatch Form FDA 3500A-Mandatory Reporting, which can be obtained from the FDA website: www.fda.gov/medwatch/getforms.htm, or other Pfizer agreed-upon form for SAE reporting.

Appendix 4 Substrates, Inhibitors, and Inducers of CYP3A4 *,**

Sensitive CYP3A4 substrates as well as those with a narrow therapeutic range (indicated in boldface and/or by review with Investigational Pharmacist) are prohibited.[13]**

Sensitive CYP3A4 substrates refers to drugs whose plasma AUC values have been shown to increase 5-fold or higher when co-administered with a known CYP3A4 inhibitor. CYP3A4 substrates with narrow therapeutic range refers to drugs whose exposure-response indicates that increases in their exposure levels by the concomitant use of CYP3A4 inhibitors may lead to serious safety concerns (such as torsades de pointes). Other CYP3A4 substrates should be avoided or used with caution.

Strong CYP3A4 inhibitors (indicated in boldface and/or by review with Investigational Pharmacist) are prohibited.

Moderate inhibitors (indicated in underlined text) and *weak inhibitors* (indicated in italicized text) should be avoided, if possible, and their concomitant use must be evaluated by the Study Chair or Investigational Pharmacist. A strong inhibitor is one that caused a ≥ 5-fold increase in the plasma AUC values or more than 80% decrease in clearance of CYP3A4 substrates (not limited to midazolam, a sensitive CYP3A4 substrate) in clinical evaluations. A moderate inhibitor is one that caused a ≥ 2- but < 5-fold increase in the AUC values or 50-80% decrease in clearance of sensitive CYP3A substrates when the inhibitor was given at the highest approved dose and the shortest dosing interval in clinical evaluations. A weak inhibitor is one that caused a ≥ 1.25 - but < 2-fold increase in the AUC values or 20-50% decrease in clearance of sensitive CYP3A substrates when the inhibitor was given at the highest approved dose and the shortest dosing interval in clinical evaluations.

All CYP3A4 Inducers (indicated in boldface and/or by review with Investigational Pharmacist) are prohibited.

Please note that these lists are not exhaustive, and there may be some cases where a boldface drug does not pose significant risk for interaction and may be used with caution (depending on dose, interval, mode of administration, etc.). If investigators are unsure of potential for drug-drug interaction, the Investigational Pharmacist and Study Chair will be consulted to determine if an agent may be used with caution versus discontinued_For the most up to date table of cytochrome P450-related drug interactions, please refer to the Indiana University drug interaction table at the following website:

http://medicine.iupui.edu/clinpharm/ddis/ClinicalTable.asp.

CYP3A4,5,7 Substrates

Macrolide antibiotics: Clarithromycin Erythromycin (not 3A5) NOT azithromycin Telithromycin Anti-arrhythmics: **Quinidine 3OH** (not 3A5) Benzodiazepines: Alprazolam Diazepam 3OH	Miscellaneous: **Alfentanyl** Aprepitant Aripiprazole **Budesonide** **Buspirone** Cafergot Caffeine_TMU Cilostazol Cocaine Codeine-Ndemethylation

CYP3A4,5,7 Substrates

Midazolam **Triazolam** Immune Modulators: **Cyclosporine** **Sirolimus** **Tacrolimus (FK506)** HIV Antivirals: Indinavir Nelfinavir Ritonavir **Saquinavir** Prokinetic: **Cisapride** Antihistamines: **Astemizole** Chlorpheniramine **Terfenadine** Calcium Channel Blockers: Amlodipine Diltiazem **Felodipine** Lercanidipine Nifedipine2 Nisoldipine Nitrendipine Verapamil HMG CoA Reductase Inhibitors: Atorvastatin Cerivastatin **Lovastatin** NOT pravastatin **Simvastatin** Steroid 6beta-OH: Estradiol Hydrocortisone Progesterone Testosterone	Dapsone Dexamethasone Dextromethorphan **Diergotamine** Docetaxel Domperidone **Eletriptan** **Eplerenone** **Ergotamine** **Fentanyl** Finasteride **Fluticasone** Gleevec Haloperidol Irinotecan LAAM Lidocaine Methadone Nateglinide Ondansetron **Pimozide** Propranolol Quetiapine Quinine Risperidone NOT rosuvastatin Salmeterol **Sildenafil** Tamoxifen Taxol **Terfenadine** Trazodone **Vardenafil** Vincristine Zaleplon Ziprasidone Zolpidem

CYP3A4,5,7 Inducers

Barbiturates	**Oxcarbazepine**
Carbamazepine	**Phenobarbital**
Efavirenz	**Phenytoin**
Glucocorticoids* **	**Pioglitazone**
Modafinil	**Rifabutin**
Nevirapine	**Rifampin**
	St. John's Wort
	Troglitazone

CYP3A4,5,7 Inhibitors

Amiodarone	**Itraconazole**
Amprenavir	**Ketoconazole**
Aprepitant	Mibefradil
Atazanavir	Mifepristone
Chloramphenicol	**Nefazodone**
Cimetidine	**Nelfinavir**
Clarithromycin	Norfloxacin
Celaviridine	Norfluoxetine
Diethyldithiocarbamate	**Ritonavir**
Diltiazem	**Saquinavir**
Erythromycin (NOT azithromycin)	Star fruit
Fluconazole	**Telithromycin**
Fluvoxamine	Verapamil
Fosamprenavir	Voriconazole
Gestodene	
Grapefruit juice	
Imatinib	
Indinavir	

* Flockhart DA. Drug Interactions: Cytochrome P450 Drug Interaction Table. Indiana University School of Medicine (2007). Http://medicine.iupui.edu/clinpharm/ddis/table.asp Accessed 6/12/2009.

**http://www.fda.gov/Drugs/DevelopmentApprovalProcess/DevelopmentResources/DrugInteractionsLabeling/ucm081177.htm#cypEnzymes. Accessed 6/17/2009.

*** Glucocorticoids are permitted for transient use such as for treatment of or premedication for hypersensitivity reactions, asthma or chronic bronchitis exacerbations, or nausea. Chronic use (≥ 2 weeks) is prohibited.

Appendix 5 Substrates of CYP2B6, CYP2C8, and CYP2C9*,**

Sensitive CYP2B6, CYP2C8, and CYP2C9 substrates as well as those with a narrow therapeutic range (indicated in boldface and/or by review with Investigational Pharmacist) are prohibited.[13] Other CYP2B6, CYP2C8, and CYP2C9 substrates should be avoided or used with caution.

Please note that these lists are not exhaustive, and there may be some cases where a boldface drug does not pose significant risk for interaction and may be used with caution (depending on dose, interval, mode of administration, etc.). If investigators are unsure of potential for drug-drug interaction, the Investigational Pharmacist and Study Chair will be consulted to determine if an agent may be used with caution versus discontinued For the most up to date table of cytochrome P450-related drug interactions, please refer to the Indiana University drug interaction table at the following website:

http://medicine.iupui.edu/clinpharm/ddis/ClinicalTable.asp

CYP2B6 Substrates	CYP2C8 Substrates	CYP2C9 Substrates
Bupropion	Amodiaquine	Celecoxib
Cyclophosphamide	Cerivastatin	Diclofenac
Efavirenz	**Paclitaxel**	Fluoxetine
Ifosfamide	**Repaglinide**	Fluvastatin
Methadone	Torsemide	Glibenclamide
		Glimepiride
		Glipizide
		Glyburide
		Ibuprofen
		Irbesartan
		Lornoxicam
		Losartan
		Meloxicam
		Nateglinide
		Phenytoin-4-OH2
		Piroxicam
		Rosiglitazone
		S-naproxen
		Suprofen
		S-warfarin
		Tamoxifen
		Tolbutamide
		Tolbutamide
		Torsemide

* Flockhart DA. Drug Interactions: Cytochrome P450 Drug Interaction Table. Indiana University School of Medicine (2007), http://medicine.iupui.edu/clinpharm/ddis/table.asp. Accessed 6/12/2009.

**http://www.fda.gov/Drugs/DevelopmentApprovalProcess/DevelopmentResources/DrugInteractionsLabeling/ucm081177.htm#cypEnzymes. Accessed 6/17/2009.

Appendix 6 Substrates of UGT1A1 and UGT1A9*

Substrates of UGT1A1 and UGT1A9 should be used with caution. If used concomitantly with sorafenib, levels of these substrates are expected to increase. Please note that these lists are not exhaustive, and there may be some cases where a boldface drug does not pose significant risk for interaction and may be used with caution (depending on dose, interval, mode of administration, etc.). If investigators are unsure of potential for drug-drug interaction, the Investigational Pharmacist and Study Chair will be consulted to determine if an agent may be used with caution versus discontinued

UGT1A1 Substrates	UGT1A9 Substrates
Acetaminophen	Acetaminophen
Buprenorphine	Ibuprofen
Ethinyl estradiol	Phenytoin
Ibuprofen	Valproate
Irinotecan	Warfarin
Nicotine	
Phenytoin	
Warfarin	

***** Adapted from GeneMedRx Health Care Provider Information, http://www.genemedrx.com/UGTtable.php. Accessed 6/12/09

Appendix 7 Substrates, Inhibitors, and Inducers of P-glycoprotein (P-gp)*

In general, inhibitors and inducers of P-gp are to be avoided or used with caution.

Use of the P-gp substrates digoxin and doxorubicin is prohibited (indicated in boldface and/or by review with Investigational Pharmacist).

Temsirolimus has been shown to inhibit P-gp mediated efflux of digoxin *in vitro*; sorafenib increases concentrations of doxorubicin. Please note that these lists are not exhaustive, and there may be some cases where a boldface drug does not pose significant risk for interaction and may be used with caution (depending on dose, interval, mode of administration, etc.). If investigators are unsure of potential for drug-drug interaction, the Investigational Pharmacist and Study Chair will be consulted to determine if an agent may be used with caution versus discontinued

P-gp Substrates	P-gp Inhibitors	P-gp Inducers
Aldosterone	Atorvastatin	Amprenavir
Amprenavir	Bromocriptine	Clotrimazole
Bilirubin	Carvedilol	Dexamethasone
Cimetidine	Cyclosporine	Indinavir
Colchicine	Erythromycin	Morphine
Cortisol	GF120918	Nelfinavir
CPT-11	Itraconazole	Phenothiazine
Cyclosporine	Ketoconazole	Retinoic acid
Dexamethasone	LY335979	Rifampin
Digoxin	Meperidine	Ritonavir
Diltiazem	Methadone	Saquinavir
Domperidone	Nelfinavir	St. John's Wort
Doxorubicin	Pentazocine	
Erythromycin	Progesterone	
Estradiol-17B-Đ-glucuronide	Quinidine	
Etoposide	Ritonavir	
Fexofenadine	Saquinavir	
Indinavir	Tamoxifen	
Itraconazole	Valspodar	
Ivermectin	Verapamil	
Loperamide		
Methylprednisolone		
Morphine		
Nelfinavir		
Paclitaxel		
Quinidine		
Ranitidine		
Rhodamine		
Saquinavir		
Sparfloxacin		
Terfenadine		
Tetracycline		
Vecuronium		
Verapamil		
Vinblastine		

* Kim RB: Drugs as P-Glycoprotein Substrates, Inhibitors, and Inducers. Drug Metabolism Reviews 34 (1&2), 47-54 (2002)

Appendix 8 List of Phosphorus-Rich Foods

Below is a list of common foods that are high in phosphorus.

Please also refer to USDA National Nutrient Database for a list of foods sorted by their elemental phosphorus content:

http://www.nal.usda.gov/fnic/foodcomp/Data/SR17/wtrank/sr17a305.pdf

Phosphorus

Phosphorus is a mineral that is found in many foods. It is an essential nutrient that is present in all body tissues and is necessary for proper nerve and muscle function as well as bone health.

Phosphorus Content of Selected Foods

Grains and Cereals

1/2 cup Bran Cereal	315	mg
1 slice Cornbread	178	mg
1 Whole Wheat Pita	115	mg
1/2 cup Cooked Oatmeal	88	mg
1/2 cup Cooked Brown Rice	81	mg
1 cup Cooked Pasta	80	mg
1 slice Multi Grain Bread	65	mg
1 slice 100% Whole Wheat Bread	57	mg

Beans and Legumes

1/2 cup Cooked Lentils	178	mg
1/2 cup Cooked Pinto Beans	149	mg
1/2 cup Edamame	131	mg
1/2 cup Cooked Black-eyed Peas	119	mg
1/2 cup Cooked Kidney Beans	115	mg
1/2 cup Cooked Garbanzo Beans	108	mg
1/2 cup Cooked Split Peas	97	mg
1/2 cup Cooked Lima Beans	89	mg

Other Food Items

1 oz Chocolate	200	mg
1 tsp Brewer's Yeast	52	mg
12 oz Cola	44	mg
3 Caramels	35	mg

Dairy Products

1 cup Yogurt	385	mg
1 cup Nonfat Milk	236	mg
1/2 cup Frozen Yogurt	177	mg
1/2 cup Cottage Cheese	151	mg
1 cup Soy Milk	151	mg
1 oz Cheese	150	mg
1 cup Nondairy Creamer	132	mg
1/4 cup Soy Cheese	125	mg
1 cup Soy Yogurt	100	mg
1/2 cup Pudding	91	mg
1/2 cup Soy Ice Cream	79	mg
1/2 cup Ice Cream	70	mg

Meat and Protein

3 oz Sardines	417	mg
3 oz Organ Meats	344	mg
3 oz Salmon	252	mg
3 oz Pork Tenderloin	184	mg
3 oz Ground Beef	173	mg
3 oz Chicken	155	mg
1 oz Almonds	134	mg
3 oz Oysters	118	mg
2 Tbsp Peanut Butter	115	mg
1 Egg (whole)	104	mg
1 oz Walnuts	98	mg
3 oz Tofu	76	mg

To find the amount of phosphorus in the foods that you typically eat, use the nutrient calculator at: http://www.nal.usda.gov/fnic/foodcomp/search/

Phosphorus 4/2008