PROTOCOL SUBMISSION FORM **Amendment Form**	PROTOCOL NO. 04C0262-H	PRINCIPAL INVESTIGATOR (NIH Employee Name, Inst/Br, Telephone and e-mail): William Dahut NCI MOB 301.435.8183 dahutw@mail.nih.gov

PROTOCOL TITLE: "A Phase II Study of BAY 43-9006 (Sorafenib) in Metastatic, Androgen-Independent Prostate Cancer"

ABBREVIATED TITLE: Phase II: BAY 43-9006 in AIPC Reference Number: 303395

Amendment Version Date: 11/18/2008 Amendment Letter: I

The Following revisions were incorporated into this protocol and approved by:

- [X] Expedited Review (risk/ benefit ratio not changed)
- [ ] Full Board Review (meeting date)

Amendment includes changes required by:

- [X] N/A
- [ ] NCI IRB
- [ ] CTEP
- [ ] Other Sponsor
- [ ] FDA
- [ ] Other

If Other, list:

Amendment required Scientific Review?

- [ ] Yes
- [X] No

ADMINISTRATIVE CHANGES

- [ ] Protocol Title/ Abbreviated Title
- [ ] New Principal Investigator
- [X] NIH Personnel Change
- [ ] Non-NIH Personnel Change
- [ ] Converting to multi-institutional trial

DEC clearance required? [ ] Yes [X] No

Date submitted to IC DEC: N/A Date cleared by IC DEC: N/A

PROTOCOL CONTENT CHANGES

- [ ] Precis
- [ ] Study Objectives
- [ ] Background and Rationale
- [ ] Eligibility Assessment and Enrollment
- [ ] Implementation of Study Design
- [ ] Supportive Care
- [ ] Accrual Ceiling Changed to: N/A
- [ ] Data Collection/Evaluation
- [ ] Human Subject Protections
- [ ] Data Reporting
- [ ] IND/IDE Information
- [ ] Pharmaceutical Information
- [ ] Appendices

Does the amendment impact the risk/benefit assessment?

- [ ] Yes
- [X] No

INFORMED CONSENT DOCUMENTATION

- [ ] Text Revisions to Consent(s)
- [ ] Investigator Contact Information on Consent(s)
- [X] No Changes to Consent Form

SIGNATURE

William Dahut - applied signature on 11/18/2008 8:13 AM EST
New and Old Principal Investigators - electronic signature and date

APPROVALS

Giuseppe Giaccone - applied signature on 11/18/2008 9:04 AM EST
Branch Chief - electronic signature and date

Carvn Steakley - applied signature on 11/25/2008 10:17 AM EST
Clinical Director- electronic signature and date

Wanda Salzer - applied signature on 11/22/2008 12:40 PM EST
Chair, IRB Review - electronic signature and date

Brian Bogus 12/8/08

IRB Meeting Date:

NCI Protocol #:	
CC Protocol #: 04-C-0262 (Version I)	**CTEP # 6594**

A Phase II Study of BAY 43-9006 (Sorafenib) in Metastatic, Androgen-Independent Prostate Cancer

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Drug Sponsor: CTEP

NCI Supplied Agent: BAY 43-9006 (NSC724772)

Protocol Version Date: **11/18/2008**

BAY43-9006 IND #: 38583

Prior to Day 0	Baseline CT Scan (chest, abdomen, pelvis) Baseline bone scan Baseline electrocardiogram (within 30 days of enrollment) Baseline PSA Baseline laboratory studies (chemistry, blood profile) H&P Pathologic confirmation Tumor biopsy and/or bone marrow biopsy and aspiration
C1D1-C1D28	Begin BAY 43-9006 400 mg bid Begin PK studies
C2D1	Return to Clinic H&P PSA #2 Tumor biopsy and/or bone marrow biopsy and aspiration #2 prior Start cycle 2 day 1 BAY 43-9006
C2D1-C2D28	Continue BAY 43-9006 400 mg bid
C3D1 prior to therapy	Return to clinic Restaging CT Scan (Chest/Abdomen/Pelvis) Restaging Bone scan PSA #3 If no evidence of progressive disease, begin C3D1

PRÈCIS

BAY 43-9006 (sorafenib) is a potent inhibitor of wild-type and mutant b-Raf and c-Raf kinase isoforms in vitro. In addition, this agent also inhibits p38, c-kit, VEGFR-2 and PDGFR-β affecting tumor growth as well as possibly promoting apoptosis by events downstream of c-Raf. At this time, over 500 patients have been treated with this drug with tolerable side effects.

The primary objective of this study is to determine if BAY 43-9006 is associated with a 50% 4 month probability of progression free survival in patients with metastatic AIPC as determined by clinical, radiographic, and PSA criteria.

The secondary objective of this study will be demonstration of biologic effect by the drug in the patient and on the tumor (when possible). Correlative studies will be conducted on serially obtained tissue biopsies, bone marrow biopsies, and white blood cell collections. These laboratory correlates will include elucidation of activation of components of the Raf-ERK-MEK and angiogenesis pathways using protein microarray technologies developed by the NCI/FDA clinical proteomics program.

Per Amendment D, a secondary objective of this study will also be to determine the time to progression measured by clinical and radiographic criteria. The 22 patients treated on the first stage of this protocol will be retrospectively evaluated with respect to this secondary endpoint if possible. Please refer to the statistics section for further details.

The combination of correlated clinical and laboratory endpoints with emphasis on molecular signaling will provide new information on the anti-tumor effects helping to characterize its role in the treatment of AIPC.

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1. OBJECTIVES

1.1 Primary Objective

The primary objective of this study is to determine if BAY 43-9006 is associated with a 50% 4 month probability of progression free survival in patients with metastatic AIPC as determined by clinical, radiographic and PSA criteria.

1.2 Secondary Objectives

- Measurement of overall response rate and overall survival
- Per Amendment D, a secondary objective of this study is to determine whether BAY 43-9006, when used to treat metastatic AIPC, is associated with ≥50% of patients progression free at 4 months by clinical and radiographic criteria. The 22 patients treated on the first stage of this protocol will be retrospectively evaluated with respect to this secondary endpoint. Please refer to the statistics section for further details.
- Demonstration of biologic effect by BAY 43-9006 in the patient and on the tumor (when possible) via correlative studies will be conducted on serially obtained tissue biopsies, bone marrow biopsies, and white blood cell collections. Laboratory correlates will include elucidation of activation of components of the Raf-ERK-MEK and angiogenesis pathways using protein microarray technologies developed by the NCI/FDA clinical proteomics program.
- Measurement of the pharmacokinetics of BAY 43-9006 in human patients with prostate cancer.
- Description of the prostate specific antigen (PSA) response rate to therapy with BAY 43-9006.

2. BACKGROUND

2.1 Androgen-Independent Prostate Cancer (AIPC)

Prostate adenocarcinoma is the most common malignancy in American men and the second leading cause of cancer related deaths [1, 2]. According to the SEER database, 42% of patients have either metastatic disease or will eventually progress following local therapy. Whereas androgen ablation therapy is an effective initial modality, androgen independence and progression of disease eventually occurred in all patients with metastatic prostate cancer [3, 4]. The utilization of second line hormonal agents is generally associated with low response rates, and has no documented survival benefit.

Chemotherapies have been extensively evaluated in patients with metastatic androgen independent prostate cancer (AIPC) since the 1970s. The initial studies showed low response rates and high toxicities. Recently, however, with the development of new agents targeting prostate cancer both on the cellular and molecular level, promising results have been emerging. The agents, including docetaxel, mitoxantrone, estramustine, vinblastine and etoposide, either as a single agent or as a combination therapy, have produced significant PSA responses in 30-80% of patients with median progression free survival of 3-5 months [5]. Chemotherapeutic agents also showed benefits in pain control, and/or quality of life, with estramustine/docetaxel combination showing the most promise. In our prior trial of thalidomide in a similar patient population, the median time to treatment failure was 2.2 months. However, the benefit in overall survival is still unknown. There is no standard therapy for patients in this population which has been shown to have an impact on survival. Therefore, new therapeutic modalities are needed in patients with advanced AIPC.

2.2 BAY 43-9006 (Sorafenib)

BAY 43-9006 represents a novel class of anticancer agents known as bi-aryl ureas. It is a potent inhibitor of wild-type and mutant b-Raf and c-Raf kinase isoforms in vitro. In addition, this agent also inhibits p38, c-kit, VEGFR-2 and PDGFR-β affecting tumor growth as well as possibly promoting apoptosis by events downstream of c-Raf. At this time, over 500 patients have been treated with this drug with tolerable side effects [6].

Activation of the *Ras* oncogene signaling pathway is considered to be an important mechanism by which human cancer develops. Raf kinase is a protein involved in the Ras signal transduction pathway. Ras regulates several pathways which synergistically induce cellular transformation, including the

Raf/MEK/ERK cascade and the rac and rho pathways [7, 8]. In particular, Ras activates the Raf/MEK pathway by first localizing Raf to the plasma membrane, where Raf initiates a mitogenic kinase cascade. Activated Raf phosphorylates and activates MEK which in turn phosphorylates and activates ERK. Activated ERK then translocates from the cytoplasm into the nucleus and modulates gene expression via the phosphorylation of transcription factors. Thus activation of Raf kinase, via activation of Ras, is thought to play an important role in carcinogenesis.

In particular, B-Raf, a serine/threonine kinase, has been shown to be activated in a number of human tumor types including melanoma, ovarian and papillary thyroid carcinomas [9-15]. A survey of 43 cancer cell lines showed that all B-Raf mutations resided in exons 11 or 15. Remarkably, 80% of these B-Raf mutations represent a single nucleotide change of T-A at nucleotide 1796 resulting in a valine to glutamic acid change at residue 599 (V599E, exon 15) in the CR3 domain (ATP binding and substrate recognition) which in turn confers constitutive kinase activity [10, 11].

***In Vitro* Activity**

The ability of BAY 43-9006 to inhibit a number of kinases was evaluated [6]. The *in vitro* biochemical and cellular profile of BAY 43-9006 is summarized below:

Biochemical Assay	**IC_{50} (μM)**
c-Raf[b]	0.002/0.006
b-Raf wild-type	0.025
b-Raf V599E mutant	0.038
VEGFR-2 (human)	0.090
VEGFR-2 (murine)	0.006
VEGFR-3 (murine)	0.010
PDGFR-β (murine)	0.028
Flt-3	0.058
c-KIT	0.068
FGFR-1	0.580
p38α	0.038
Cellular Mechanism[c]	**IC_{50} (μM)**
MDA-MB-231 MEK phosphorylation (Human Breast)	0.04
BxPC-3 MEK phosphorylation (Human Pancreatic)	1.00
LOX ERK phosphorylation (Human Melanoma)	0.80
b-Raf ER MEK activation (Human Chimera, 3T3 cells)	2.30
VEGFR-2 phosphorylation (Human, 3T3 cells)	0.03
VEGFR-3 phosphorylation (Mouse, 293 cells)	0.10
PDGFR-β phosphorylation (Human, AoSMC)[d]	0.02
Cellular Proliferation	**IC_{50} (μM)**
MDA-MB-231 (10% FCS)[e]	2.60
MDA-MB-231 (0.1% FCS)	0.10
VEGF-HUVEC (2.0% FCS)[f]	3.00

PDGFR-β AoSMC[d] (0.1% BSA)[g]	0.23

a Recombinant enzyme assay

b Raf kinase activated with Lck (truncated/full length c-Raf)

c Mechanistic cellular assays all performed in 0.1% BSA

d Human aortic smooth muscle cells

e Fetal calf serum

f Human umbilical vein endothelial cells

g Bovine serum albumin

In vitro kinase assays demonstrated that BAY 43-9006 is a potent inhibitor of wildtype and mutant (V599E) B-Raf and c-Raf Kinase isoforms *in vitro* [6]. In addition, BAY 43-9006 did not inhibit human EGFR or Her2 kinases at 10 μM. Nor were PKC-α, PKC-β, PKC-γ, and PKA (rat, rabbit and bovine sources) kinase activity inhibited *in vitro*. BAY 43-9006 demonstrated an IC_{50} of 780 nM against p59 (bovine) Fyn kinase (Src family of protein tyrosine kinases). In non-kinase targets BAY 43-9006 had moderate potency against the adenosine A3, dopamine D1, and muscarinic M3 receptors with IC_{50} of 1.6 μM, 2.0 μM, and 3.1 μM, respectively. BAY 43-9006 did not inhibit MEK-1, ERK-1, EGFR, HER2/neu, c-met, PKA, PKB, Cdk-1/cyclin B, pim-1, GSK 3-b, CK-2, PKC-α (r), PKC-β (r), PKC-γ at concentrations as high as 10 μM. In summary, BAY 43-9006 showed ≥100-fold more selectivity for Raf kinase relative to other target proteins.

BAY 43-9006 also inhibited *in vitro* several receptor tyrosine kinases (RTKs) that are involved in tumor progression; human VEGFR-2, murine VEGFR-2, murine VEGFR-3, murine PDGFR-β, Flt-3, c-KIT, and p38α (MAPK family). In cellular assays, BAY 43-9006 was found to be a potent inhibitor of human and murine VEGFR-2, murine VEGFR-3, and murine PDGFR-β receptor phosphorylation [6].

VEGF and PDGF receptors are involved in the mechanism of tumor angiogenesis [16, 17]. PDGF receptors may also play a role in patients with chronic myeloproliferative cancers [18]. Flt-3 is important in acute myelogenous leukemia [13] and c-Kit plays a critical role in gastrointestinal stromal tumors [19].

In Vivo Activity

BAY 43-9006 has demonstrated *in vivo* anti-tumor efficacy as a single agent against a broad range of human tumor xenografts as summarized in the following table. The models evaluated include HCT-116 and DLD-1 colon tumor xenografts, MX-1 mammary tumor xenograft, NCI-H460 and A549 NSCLC xenografts, MiaPaCa-2 pancreatic tumor xenografts, and SK-OV-3 ovarian tumor xenografts. In this table, compound efficacy is expressed as percent tumor growth inhibition (TGI) and is calculated as ((1-(T/C)) *100, where T and C represent the mean tumor size in the Treated and Control groups respectively at the first measurement after the end of treatment.

BAY 43-9006 Demonstrates Broad Spectrum Anti-Tumor Efficacy in Preclinical Xenograft Models

Tumor Type	Model	Dose (mg/kg/dose free base equiv.)[1]	Percent TGI ((1-(T/C))*100)
Colon	HCT-116	10	45
		30	64
		100	68
Colon	DLD-1	15	31
		30	66
		60	75
NSCLC	NCI-H460	10	27
		30	56
NSCLC	A549	30	60
		60	68
Mammary	MX-1	30	51
		60	67
Pancreatic	Mia-PaCa-2	10	45
		30	66
		100	73
Ovarian	SK-OV-3	10	58
		30	64
		100	81

[1] Compound dosed as BAY 43-9006 or equivalent dose levels of tosylate salt, BAY 54-9085

The majority of the initial anti-tumor efficacy evaluations *in vivo* were conducted in the HCT116 colon tumor model since the tumorigenicity of this cell line was previously shown to be dependent on K-Ras activation. Additional studies indicated that prolonged anti-tumor efficacy could be attained by extending the duration of treatment and that, in this tumor model, BAY 43-9006 was able to arrest tumor growth even if therapy was initiated against a substantially greater tumor burden.

BAY 43-9006 also showed significant oral activity against two additional human tumor xenograft models that contain K-Ras mutations: MiaPaCa-2 pancreatic carcinoma and H460 non-small cell lung carcinoma. The anti-tumor efficacy of BAY 43-9006 was also evaluated against the human SKOV-3 ovarian tumor cell line that contains a wild-type Ras but over-expresses both the EGF and Her2 growth factor receptors. These receptors also signal through the Ras/Raf/MEK pathway.

In human tumor xenografts, MDA-MB-231 (breast) and Colo-205 (colon), there was a dramatic reduction of tumor neo-vascularization [6]. Recent data also indicated that inhibition of c-Raf may promote cell death in endothelial cells as a downstream event of VEGFR-2 stimulation [20].

Taken together, data suggests that BAY 43-9006 may be of therapeutic value not only in human tumors containing *Ras* gene mutations, but also in tumors over-expressing growth factor receptors in the Ras/Raf/MEK pathway, and by inhibiting tumor angiogenesis or neo-vascularization through inhibition of VEGFR-2, VEGFR-3, and/or PDGFR-β.

The ability of BAY 43-9006 (or its tosylate salt, BAY 54-9085) to be combined with paclitaxel, irinotecan, gemcitabine, or cisplatin was evaluated in preclinical *in vivo* models. In these studies, the focus was to evaluate if the co-administration of BAY 43-9006 would adversely affect the tolerance or anti-tumor efficacy of the 'standard of care' agent. The general health of mice was monitored and mortality was recorded daily. Tumor dimensions and body weights were recorded twice a week starting with the first day of treatment. Treatments producing greater than 20% lethality and/or 20% net body weight loss were considered 'toxic'. The results of these combinability analyses are summarized below:

Combinability of Concurrent Treatment with BAY 43-9006 and Clinically Established Agents

Combination Agent	Tumor Model	Combinability Y/N
Paclitaxel	NCI-H460 NSCLC	Yes
	MX-1 Mammary	Yes
Irinotecan	DLD-1 Colon	Yes
Gemcitabine	MiaPaCa-2 Pancreatic	Yes
Cisplatin	NCI-H23 NSCLC	Yes

BAY 43-9006 can be safely combined with a variety of standard cytotoxic cancer chemotherapy agents, including paclitaxel, irinotecan, gemcitabine and cisplatin with no significant increase in the toxicity associated with those agents and without diminishing their anti-tumor efficacy in preclinical models.

Clinical Experience

BAY 43-9006 has been evaluated in multiple Phase 1 and Phase 2 studies in a variety of tumor types. To date, over 500 patients have been treated with single agent BAY 43-9006. The Phase 1 single agent clinical plan has focused on characterizing the safety and pharmacokinetic profile BAY 43-9006 in several different dosing regimens. All Phase 1 patients had a variety of advanced refractory solid tumors, and some of the patients stayed on trial for more than one year. Four different regimens have been tested: continuous treatment, 4 weeks on/ 1 week off, 3 weeks on /1 week off, and 1 week on/ 1 week off. Patients have received doses ranging from 50 mg once weekly to 1600 mg daily of BAY 43-9006 on intermittent and continuous schedules. The 800 mg bid continuous administration cohort has exceeded maximum tolerated dose (MTD) in all tested schedules. The 600 mg bid cohort exceeded the MTD in all but the less dose intensive regimen of 1 week on / 1 week off. The most frequent drug-related adverse events were hand-foot skin reaction, dermatitis, rash, fatigue, anorexia and diarrhea. The most common dose limiting toxicities (DLTs) in the phase I trials were hand-foot syndrome, fatigue, pain, diarrhea, elevated alkaline phosphatase, and abdominal pain/cramping. DLTs were rarely attriubuted to the following: hypertension weight loss, anorexia, pancreatitis, elevated amylase and lipase without pancreatitis,

There was an increase in the number of serious adverse events, discontinuations due to adverse events, and a number of skin toxicities at the higher dose levels ≥ 600 mg bid. Therefore, 400 mg bid was selected as the recommended dose for Phase 2.

Currently, the Phase 2 program includes studies designed to explore anti-tumor efficacy in certain tumor types and to gain additional experience with pharmacokinetics and safety. Thus far, Phase 2 studies have enrolled over 300 patients with a variety of tumor types including colorectal, renal cell, hepatocellular, pancreatic, and thyroid cancer and melanoma as well as several less common tumors.

In general, available information from the ongoing Phase 2 studies reveal toxicities that are similar to the Phase 1 data. Again, the five most frequent drug-related toxicities observed include hand-foot skin

reaction, rash, anorexia, diarrhea, and fatigue. When all available data from the various studies/schedules are combined, the incidence of greater than grade 3 treatment emergent skin toxicity (e.g. hand-foot syndrome and “dermatology/skin reaction”) for an initial dose of 400 mg bid and 600 mg bid, was 0% and 30%, respectively. Anti-tumor activity was observed in both Phase 1 and 2 studies.

Pharmacokinetics

Pharmacokinetic studies in humans shows that BAY 43-9006 undergoes metabolism to a few primary metabolites, some of which may be active [6]. No data are available at this writing to describe the pharmacokinetics of repetitive dosing of BAY 43-9006 in patients**.** However, up to 36 hours after a single dose, BAY 43-9006 continues to be the predominant species detected. In vitro liver microsomal degradation studies data indicate that BAY 43-90006 is primarily metabolized by CYP 3A4. Ongoing studies are being pursued by the Sponsor to evaluate drug-drug interaction potential of BAY 43-9006 with agents that alter function of CYP2C19, CYP2D6, and CYP3A4. Moderate fat meals do not affect bioavailability of BAY 43-9006; it is in fact recommended to be administered with or around meals. Approximately 19% of dose is excreted in urine and 76% in feces.

2.3 Rationale

Specific to prostatic adenocarcinomas, strong epithelial staining for Ras oncoproteins has been seen with a significant difference in expression levels between normal and tumor cells. Furthermore, an inverse correlation between Ras positivity and degree of differentiation and survival has been reported [21, 22]. Such findings suggest that functional activation of wild-type Ras might contribute to prostate tumorigenesis.

Bakin and colleagues [23, 24] demonstrated that manipulation of Ras-related signaling affected sensitivity to hormonal interventions in vitro. This pathway is involved in the mediation of responses to a number of growth factors as well angiogenic factors. More recently, Fu and colleagues have been able to demonstrate that when Raf kinase inhibitor protein (RKIP) is transfected into C4-2B cells (malignant prostatic rat epithelial cells derived from LNCaP cells with a metastatic phenotype), invasion was decreased and development of lung metastases in animal models was no longer seen suggesting that this pathway is associated with suppression of tumor metastases [25].

In addition to activity on Raf-associated signaling, BAY 43-9006 has been described to affect human VEGFR-2, murine VEGFR-2, murine VEGFR-3, murine PDGFR-β, Flt-3, c-KIT, and p38α (MAPK family). Multiple reports in the available literature report VEGF expression in most prostatic carcinomas [26] and expression of VEGFR may relate to migratory and invasive ability. Furthermore, in vitro models have suggested that activation of p38-MAPK active pro-apoptotic pathway activity in prostate cancer [27]. While the remainder of the targets listed above have not been well characterized in prostate cancer, their impact on the angiogenic cascade is of interest in prostate cancer as several groups have proposed manipulation of angiogenic signaling as a therapeutic target [28].

Given the potential role of the Ras-Raf pathway in metastatic prostate cancer in addition to the known in vivo and in vitro activity of the agent, we propose the execution of a phase II trial to measure the efficacy of this compound in preventing progression of AIPC. At this time little data exists for this agent in AIPC. This study represents the first formal inquiry into this matter.

3. PATIENT SELECTION

3.1 Eligibility Criteria

3.1.1 Patients must have histopathological confirmation of prostate cancer by the Laboratory of Pathology of the NCI or Pathology Department of the National Naval Medical Center prior to entering this study. Patients whose pathology specimens are no longer available may be enrolled in the trial if the patient has a clinical course consistent with prostate cancer and available documentation from an outside pathology laboratory of the diagnosis. In cases where original tissue blocks or archival biopsy material is available, all efforts should be made to have the material forwarded to the research team for use in correlative studies.

3.1.2 Patients must have metastatic progressive androgen-independent prostate cancer. There must

be radiographic evidence of disease after primary treatment with either surgery or radiotherapy that has continued to progress despite hormonal agents. Progression requires that a measurable lesion is expanding, new lesions have appeared, and/or that PSA is continuing to rise on successive measurements. Patients on flutamide must have disease progression at least 4 weeks after withdrawal. Patients on bicalutamide or nilutamide must have progression at least 6 weeks after withdrawal.

3.1.3 Patients may have had no more than 1 previous cytoxic chemotherapeutic line.

3.1.4 Because no dosing or adverse event data are currently available on the use of BAY 43-9006 in patients <18 years of age, children are excluded from this study.

3.1.5 Patients must have a life expectancy of more than 3 months.

3.1.6 Patients must have a performance status of 0 to 2 according to the ECOG criteria.

3.1.7 Patient must have adequate organ function as defined below:

Leukocytes	≥3,000/µl
absolute neutrophil count	≥1,500/µl
Platelets	≥100,000/µl
total bilirubin	≤1.5 X institutional upper limits of normal
AST(SGOT) and ALT(SGPT)	≤2.5 X institutional upper limit of normal
creatinine OR Creatinine clearance	≤1.5 X institutional upper limits of normal ≥60 mL/min/1.73 m^2 for patients with creatinine levels above institutional normal.

3.1.8 Patients must have recovered from any acute toxicity related to prior therapy, including surgery. Toxicity should be ≤ grade 1 or returned to baseline.

3.1.9 Hormonal profile: all patients who have not undergone bilateral surgical castration must continue suppression of testosterone production by appropriate usage of GnRH agonists.

3.1.10 Patients must not have other invasive malignancies (within the past two years with the exception of non-melanoma skin cancers or non-invasive bladder cancer).

3.1.11 Patients must be able to understand and sign an informed consent form.

3.1.12 Concurrent use of bisphosphonates will be allowed for patients with known bone metastases

3.1.13 Patients who require hematopoietic growth factor support (e.g. epogen, darbepoetin) , NSAIDs, and other maintenance medications prior to study entry will be allowed to continue their supportive therapies.

3.1.14 The effects of BAY 43-9006 on the developing human fetus at the recommended therapeutic dose are unknown. For this reason and because Raf-kinase inhibitors are known to be teratogenic, men must agree to use adequate contraception (abstinence; hormonal or barrier method of birth control) prior to study entry and for the duration of study participation.

3.2 **Exclusion Criteria**

3.2.1 Patients who have had chemotherapy or radiotherapy (including radioisotopes) within 4 weeks (6 weeks for nitrosoureas or mitomycin C; >3 months for suramin) prior to entering the study or those who have not recovered from adverse events due to agents administered more than 4 weeks earlier.

3.2.2 Patients may not be receiving any other investigational agents.

3.2.3 Patients with known brain metastases are excluded from this clinical trial because of their poor

prognosis and because they often develop progressive neurologic dysfunction that would confound the evaluation of neurologic and other adverse events.

3.2.4 History of allergic reactions attributed to compounds of similar chemical or biologic composition to BAY 43-9006

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

3.2.6 Hypertension as defined by systolic blood pressure in excess of 170 mmHg or diastolic pressure in excess of 100 mmHg. Patients with a history of hypertension that is well controlled on medication will not be excluded. Patients may not be on either verapamil or diltiazem while on study. Use of calcium channel blocker should be discouraged.

3.2.7 Patients with immune deficiency are at increased risk of lethal infections when treated with marrow-suppressive therapy. Therefore, HIV-positive patients receiving combination anti-retroviral therapy are excluded from the study because of possible pharmacokinetic interactions with BAY 43-9006. Appropriate studies will be undertaken in patients receiving combination anti-retroviral therapy when indicated.

3.2.8 History of bleeding diathesis

3.2.9 Patients on therapeutic anticoagulation are at increased risk from bleeding while on BAY 43-9006. Prophylactic anticoagulation (i.e. low dose warfarin) of venous or arterial access devices is allowed provided that the requirements for PT, INR or PTT are met: PT < 1.1 x institutional upper limit of normal; INR < 1.1; PTT within the institutional limits of normal. These requirements will only be made upon those patients on warfarin.

3.3 Inclusion of Women and Minorities

Men of all races and ethnic groups are eligible for this trial. Women are excluded by the nature of the disease.

3.4 Baseline evaluation

3.4.1 Imaging Studies (Baseline- obtained within one month prior to enrollment): Technetium-99 Bone Scintigraphy, Chest X-ray, CT scan of chest, abdomen and pelvis

3.4.2 Laboratory Evaluation (Baseline- obtained within 16 days prior to enrollment)

3.4.2.1 Hematological Profile: CBC with differential and platelet count, PT, aPTT, fibrinogen

3.4.2.2 Biochemical Profile: electrolytes, BUN, creatinine, AST, ALT, total bilirubin, calcium, phosphorus, albumin, magnesium, uric acid, C-reaction protein, albumin, amylase, lipase, testosterone level.

3.4.2.3 Tumor Marker Profile: PSA (baseline within 7 days prior to enrollment), acid phosphatase (if PSA <4 ng/mL, for research purposes only)

3.4.2.4 Electrocardiogram (performed within 30 days prior to enrollment); Patients with a history of myocardial infarction or cardiac arrhythmias will undergo evaluation by cardiology.

3.5 Patient Registration

3.5.1 Authorized physicians or designees must telephone information concerning an eligible candidate to the Orkand personnel (Orkand is the current data management contract company for the Center for Cancer Research of the NCI) by fax at (301) 480-0757 between the hours of 8:30am and 5:00pm, Monday through Friday.

3.5.2 At the time of enrollment, eligibility criteria will be queried. Protocol specific eligibility checklist will be developed by the Orkand staff and reviewed by the principal investigator.

3.5.3 The Orkand personnel are to be called at the time any patient is taken off the study

4. TREATMENT PLAN

This study will be executed in a two-stage optimal design as described by Simon et al [29]. In the first stage, 22 patients will be enrolled and evaluated for progression. If 8 patients show disease stability beyond 4 months accrual will continue to the full 46 patients. If among the first 22 patients, 7 or fewer are able to be progression free at the 4 month evaluation, then no further patients will be enrolled and the study will close to accrual. Should 18 or more patients remain progression free at 4 months, BAY 43-9006 will be considered worthy of further development in clinical trials. If 8-17 patients are progression free at 4 months, the study set will be considered insufficient. Under the null hypothesis (30% progression free at 4 months), the probability of being able to stop accrual after 22 patients have been evaluated at 4 months is 67%.

The study was originally designed to have a two-stage design with 22 and then up to a total of 46 patients evaluating progression using PSA criteria. The results from the first stage were inadequate to justify accrual to the second stage. However, per amendment D, waiver of the stopping rule is requested to allow for accrual to the second stage.

4.1 BAY 43-9006 (Sorafenib) Administration

At the outset of the study, the patient will be admitted to the inpatient service for a period of approximately 24 hours to complete research studies including biopsies and PK measurements. Otherwise, treatment will be administered on an outpatient basis. Reported adverse events and potential risks are described in Section 6. Appropriate dose modifications for BAY 43-9006 are described in Section 5. No investigational or commercial agents or therapies other than those described below may be administered with the intent to treat the patient's malignancy.

BAY 43-9006 is supplied as 200-mg tablets and is administered orally twice a day. Patients are to swallow the tablets whole with approximately 250 ml (8 oz.) of water, each morning and evening (i.e., 12-hourly). Tablets may be taken with or without food. BAY 43-9006 will be given as self-administered oral doses of 400 mg bid continuously throughout the 28 day cycle. Patients will be asked to administer the drug at approximately 08:00 and 20:00 hours. Patients will be asked to maintain a diary to document consumption of BAY 43-9006 as out lined in section 6 and 12.6.7.

Patients will be required to have weekly blood pressure monitoring for the first 4 weeks of the study at a healthcare facility given the risk of hypertension from BAY 43-9006. This will be logged into the patient's study diary during the first 4 weeks of treatment.

4.2 Supportive Care Guidelines

4.2.1 No premedications will be used with the initiation of therapy.

4.2.2 Nausea

Should patients develop persistent nausea, they will be given appropriate antiemetic therapy following clinical center guidelines starting with prochlorperazine 10 mg PO/IV q8h prn.

4.2.3 Hand-Foot Syndrome

- Treatment with topical emollients (such as Aquaphor) for symptom relief is permitted.
- Topical and/or oral steroids or antihistamine agents may be used.
- Vitamin B6 (pyridoxine; 50 - 150 mg orally each day) may also be used.

4.3 Duration of Therapy

In the absence of treatment delays due to adverse event(s), treatment may continue until one of the following criteria applies:

- Disease progression

- Intercurrent illness that prevents further administration of treatment
- Unacceptable adverse event(s)
- Patient decides to withdraw from the study, or
- General or specific changes in the patient's condition render the patient unacceptable for further treatment in the judgment of the investigator.
- Subject death

5. DOSING DELAYS/DOSE MODIFICATIONS

Toxicities will be described using CTCAE v3. The following adjustment will only apply if the toxicities reported are attributed by the investigator to be related to BAY 43-9006 therapy.

For Grade 1 toxicity, treatment with BAY 43-9006 will not be interrupted. For symptoms that last more than 7 days and have been found to be intolerable to the patient, the dose of BAY 43-9006 may be reduced by 200 mg/day (i.e.200 mg qAM and 400 mg qPM as the initial reduction).

For Grade 2 toxicity

For nausea, vomiting and diarrhea, maintain dosing with symptomatic treatment. The dose of BAY 43-9006 may be repeated if emesis occurs within 30 minutes of taking the tablet(s) OR all the tablets are seen in the emesis.

For persistent nausea, vomiting or diarrhea despite symptomatic treatment that remains unacceptable (intolerable) to the patient, reduce dose by 200 mg/day (i.e.200 mg qAM and 400 mg qPM as the initial reduction).

For other grade 2 toxicities, the dose does not need to be reduced unless side effects become intolerable to the patient.

If patients are receiving 200 mg bid at the time of occurrence/recurrence of toxicity, change the schedule of administration to 200 mg qd. Reductions below this dose/schedule will not be allowed.

Patients with intolerable or limiting toxicity while taking 200 mg qd will be removed from study. .

For Grade 3 clinical toxicity, or Grade 3/Grade 4 metabolic/hematologic toxicityHold BAY 43-9006 and reevaluate the patient at least weekly until toxicity improves to ≤ Grade 1 or pre-treatment baseline. Grade 3 metabolic toxicities which can be corrected to ≤ Grade 1 or pre-treatment baseline within 48 hours will not necessitate a dose reduction. For Grade 4 metabolic toxicity or Grade 3/4 hematologic toxicity the dose of BAY 43-9006 will be reduced by 200 mg/day. . Treatment will be discontinued in patients who experience toxicity ≥ grade 3 or 4 that does not resolve to ≤ grade 1 or baseline within 3 weeks.

If patients are receiving 200 mg bid at the time of occurrence/recurrence of toxicity, change the schedule of administration to 200 mg qd. Reductions below this dose/schedule will not be allowed.

Patients with intolerable or limiting toxicity while taking 200 mg qd will be removed from study.

For Grade 4 clinical toxicity

Patients who had clinical grade 4 toxicity (except pulmonary embolism without significant hypoxia and hemodynamic instability) will be taken off study permanently.

5.1 Hypertension

Given that hypertension is one of the major toxicities that may be experienced, all patients should have blood pressure measured and recorded weekly during the first 4 weeks and as needed, thereafter.

Grade of Event (CTCAE v.3)	Management/ Next Dose
grade 1	No dose modification
grade 2 asymptomatic and diastolic BP < 110 mm Hg	Begin (additional) anti-hypertensive therapy and continue agent.
grade 2 symptomatic/ persistent OR diastolic BP ≥ 110 mm Hg OR grade 3	1. Agent should be held until symptoms resolve and diastolic BP ≤ 100 mm Hg; treat patient with anti-hypertensives and when BAY 43-9006 is restarted, reduce dose level as described above* 2. If diastolic BP not controlled (≤ 100) on therapy, reduce dose again**
grade 4	Discontinue protocol therapy
* May be able to resume full dose later. ** Patients requiring > 2 dose reductions should go off protocol therapy.	

Current CTCAE definitions used by CTEP:

- Grade 1: asymptomatic, transient (< 24 hours) increase by > 20 mmHg (diastolic) or to
  - >150/100 if previously WNL; intervention not indicated
- Grade 2: recurrent or persistent (> 24 hours) or symptomatic increase by > 20 mmHg
  - (diastolic) or to > 150/100 if previously WNL; monotherapy may be indicated
- Grade 3: requiring more than one drug or more intensive therapy than previously
- Grade 4: life threatening (e.g. hypertensive crisis)

5.1.1 Recommended antihypertensive therapy

Patients requiring additional antihypertensive therapy while on BAY 43-9006 should be started on a beta blocker, diuretic, angiotensin converting enzyme inhibitor (ACE-I) or an angiotensin II receptor blocker (ARB). Doses should be advanced as needed until the patient is unable to tolerate the antihypertensive medication due to side effects. If further medication is required a beta-blocker should be initiated and doses advanced. Calcium channel blockers should be avoided.

5.1.2 For severe hypertension, hypertensive urgency/emergency

BAY 43-9006 should be held and the patient should be admitted. An EKG should be obtained and, if appropriate, cardiology should be consulted. Inpatient therapy for hypertension should be initiated.

5.1.3 Therapy may be continued if the hypertension is successfully controlled with additional medication. However, for hypertension that persists despite additional medical therapy or becomes symptomatic, therapy will be held as described above. If therapy is held for 4 weeks, the patient will be taken off of study.

5.2 GI Perforation (NOS)

If a patient experiences GI perforation while on study of any grade, patient must be taken off the study.

6. PHARMACEUTICAL INFORMATION

6.1 **BAY 43-9006 (Sorafenib, NSC 724772, IND 38583)**

6.1.1 Source: BAY 43-9006 will be supplied by the Bayer Health Care Corporation and distributed by CTEP

6.1.2 **Chemical name and identification**

Chemical Name: 4–-pyridine-2 carboxylic acid methylamide-4-methylbenzensulfonate.

Other Names: BAY 54-9085 is the tosylate salt of BAY 43-9006; sorafenib

Classification: Kinase inhibitor (Raf, VEGF-R, and PDGF-R)

Mechanism of Action: The Ras/Raf signaling pathway is an important mediator of responses to growth signals and angiogenic factors. This pathway is often aberrantly activated in human tumors due to presence of activated Ras, mutant b-Raf, or over expression of growth factor receptors.

BAY 43-9006 is a potent inhibitor of c-Raf, and wild-type and mutant b-Raf in vitro. Additionally, further characterization of BAY 43-9006 tosylate revealed that this agent inhibits several receptor tyrosine kinases (RTKs) that are involved in tumor progression (VEGF-R, PDGF-R, Flt3, and c-KIT) and p38α, a member of the MAPK family.

Molecular Formula: $C_{12}H_{16}ClF_3N_4O_3$ X $C_7H_8O_3S$

M.W.: BAY 43-9006 tosylate: 637 Daltons; BAY 43-9006 (free base): 465 Daltons

Approximate Solubility: 0.19 mg/100 mL in 0.1 N HCl, 453 mg/100 mL in Ethanol, and 2971 mg/100 mL in PEG 400.

How Supplied: BAY 43-9006 tosylate is supplied as an immediate-release film-coated, round, and salmon color tablet containing 200 mg of the free base, BAY 43-9006, and the excipients croscarmellose sodium, microcrystalline cellulose, hydroxypropylmethylcellulose, sodium lauryl sulfate, and magnesium stearate. The film-coat consists of hydroxypropylmethyl cellulose, polyethylene glycol, tiranium dioxide and red iron oxide. The film coating has no effect on the rate of release of the active BAY 43-9006 tosylate.

BAY 43-9006 tosylate 200 mg tablets are supplied in bottles of 140 tablets.

Storage: Store at controlled room temperature (15ºC – 25ºC). Storage conditions should not exceed 25ºC.

Stability: Stability studies with the 200 mg dosage form are ongoing. The current shelf life is 24 months when stored at controlled room temperature.

Route of Administration: Oral

Reported Adverse Events and Potential Risks:

Body as a whole:	fatigue, flu-like syndromes, fever, arthralgia
Gastrointestinal:	diarrhea, pancreatitis, elevated amylase/lipase, abdominal pain/cramping, nausea, flatulence, dyspepsia
Hepatic:	increased bilirubin, ALT, AST, GGT, LDH, and alkaline phosphatase
Metabolic and Nutritional:	anorexia
Skin:	hand-foot syndrome, characterized by palmar and plantar erythema; rash/desquamation, hypersensitivity reactions, dry skin, alopecia, nail changes, vitiligo, itching
Cardiovascular:	cardiac arrhythmias
Other:	voice changes

The following adverse events have been reported on trials but with the relationship to BAY 43-9006 still undetermined: arthritis, brain stem stroke, dyspnea, hypertension, increase PT and PTT, and weight loss.

Method of Administration: BAY 43-9006 tosylate should be taken with at least 250 mL of water and can be given without regards to meals. Food does not appear to have a

clear effect on BAY 43-9006 tosylate pharmacokinetics.

Potential Drug Interactions: BAY-9006 tosylate is metabolized by the P450 CYP3A enzyme and has been shown in preclinical studies to inhibit multiple CYP isoforms. Therefore, it is possible that BAY-9006 tosylate may interact with drugs that are metabolized by the P450 CYP isoenzymes or with drugs that inhibit CYP 3A. Close monitoring is recommended for patients taking agents with narrow therapeutic indices and metabolized by the liver, such as warfarin, phenytoin, quinidine, carbamazepine, phenobarbital, cyclosporine, and digoxin. Additionally, BAY-9006 tosylate is 97% to 99% protein bound; however, no drug interactions have been reported in studies, thus far.

Availability

BAY 43-9006 is an investigational agent supplied to investigators by the Division of Cancer Treatment and Diagnosis (DCTD), NCI.

Agent Ordering

NCI-supplied agents may be requested by the Principal Investigator (or their authorized designees) at each participating institution. Pharmaceutical Management Branch (PMB) policy requires that the agent be shipped directly to the institution where the patient is to be treated. PMB does not permit the transfer of agents between institutions (unless prior approval from PMB is obtained). Completed Clinical Drug Requests (NIH-986) should be submitted to the PMB by fax (301) 480-4612 or mailed to the Pharmaceutical Management Branch, CTEP, DCTD, NCI, 9000 Rockville Pike, EPN Rm. 7149, Bethesda, MD 20892.

Agent Accountability

The investigator, or a responsible party designated by the investigator, must maintain a careful record of the inventory and disposition of all drugs received from DCTD using the NCI Drug Accountability Record Form (DARF). BAY 43-9006, as an oral self-administered investigational agent, will be properly accounted for, handled, and disposed in accordance with CCR Policy # Clin-1 “Policy on Documenting, Handling and Disposing of Oral Investigational Agents that are Self-Administered by NCI CCR patients.” The Standard Operating Procedure # Clin-1 “Standard Operating Procedure for Conducting and Documenting Drug Accountability for Oral Investigational Agents that are Self Administered by Patients at the CCR” identifies all activities associated with drug accountability for orally self administered investigational agents. Study drug may be repackaged by the Clinical Center Pharmacy Dept. and dispensed in light-resistant HDPE containers that contain up to a 28-day supply of BAY 43-9006.. Patients will be instructed to document on a study diary the daily intake of BAY 43-9006, including the time the dose is taken, and whether or not any doses are missed. They will bring the study diary and any unused drug to their clinic appointments. Clinic staff will (1) collect all “old” [i.e., empty bottle(s), partial bottle(s) or full bottle(s)] of study drug; (2) perform a capsule count and record the results on the approved CCR Pill Count Case Report Form which is to be maintained in the research record; (3) dispense the partial and full bottle(s) of BAY 43-9006 to the patient. Unused study drug is to be returned to the research nurse who will dispose of it according to the SOP.

7. CORRELATIVE/SPECIAL STUDIES

7.1 Tissue analysis for molecular endpoints

Given the nature of this pharmacologic intervention, characterization of the actual biological manipulation will be critical in understanding the biological effects seen following administration. Tissue samples will be analyzed to achieve this goal.

The collection of correlative studies is designed to characterize a baseline molecular profile as well as a treatment-induced molecular profile. Raf mutational analysis will allow for retrospective analysis of responsiveness in the presence or absence of mutations. Proteomic analysis will provide profiles of signaling pathways pre-treatment and on treatment so as to understand how this agent affects a variety of

important signaling cascades associated with its reported molecular targets. In particular, primary endoints address key events in survival, proliferation, and angiogenesis which will affected either directly or downstream of Raf or VEGFR/PDGFR targets. Pharmacokinetic anlaysis will provide insight into dosing efficacy as it realtes to both biological activity and clinical response.

Correlative laboratory studies for this trial will be conducted in the Clinical Pharmacology Section of Dr. William D. Figg at the NCI and by Dr. Elise C. Kohn. All patients will have leukocyte collections (by standard buffy coat preparation) to evaluate for tumor and gene expression alterations related to drug therapy. Patients with accessible disease will have tumor biopsies before and during treatment for molecular analysis described below.

Tumor Biopsies	***Bone Marrow***	***Archival Tissue***	***Leukocytes***
Proteomic pathway analysis	Proteomic pathway analysis	B-Raf genomic analysis	Proteomic pathway analysis
B-Raf mutational screening	Cross-validation with tumor biopsy		Cross-validation with tumor biopsy

7.1.1 Accessible tumor biopsy

Biopsies of accessible tumors will be required prior to initiation of therapy and prior to starting cycle 2 as described. **Patients should avoid the use of aspirin or non-steroidal anti-inflammatory drugs for 1 week prior to biopsy to ensure safety.** Accessible tumors will be defined as those masses which can be safely accessed by a core needle biopsy while posing minimal risk to the patient. These lesions may be cutaneous or palpable or demonstrable under CT or ultrasound guidance. Determination of accessibility and safety will be made jointly by an investigator and a staff member from Special Procedures. Accessible tumors will be defined as those masses which can be safely accessed by a core needle biopsy under CT or ultrasound guidance or while posing minimal risk to the patient. Laser capture microdissection and cell lysates will be prepared for protein microarrays with targets as identified below. Garreth Peters (pager 104-4587-7 or 301-402-4622) or Dr. Edwin Posadas (phone 301-451-4982 pager 104-6563) should be contacted to arrange for sample collection. Samples should be preserved in OCT according to the NCI/FDA clinical proteomics program SOP (appendix B). Samples should be barcoded and stored in liquid nitrogen until the time of preparation for microdissection.

Biopsy specimens will be used for tissue lysate array analysis in the proteomics laboratory (see section 7.2) for signal pathway profiling before and during treatment as well as Raf mutation detection via TLA.

7.1.2. Bone marrow biopsy and aspiration

In addition to attempting to obtain tumor biopsies, attempts will be made to determine effect in both bone marrow and in white blood cells. Bone metastases are a common event in metastatic prostate cancer affecting about 80% of patients. In limited series, bone marrow trephines and aspirations were shown to yield metastatic tumor cells in 75% of cases using traditional histopathologic techniques [30]. Given the fact that patients often have bone as their sole site of metastasis and the relative difficulty of obtaining soft tissue biopsies for direct tumor sampling, bone marrow aspiration and biopsy may be a powerful tool in assessing biological response both in the tumor and in the tumor microenvironment. This question will be addressed using protein microarrays in conjunction with immunohistochemistry on these samples.

Marrow cores and aspirate collection will not be mandated, but will be requested of all patients. In addition, given the activity of BAY 43-9006 on angiogenic pathways and receptor tyrosine kinases, markers of angiogenesis including VEGFR and PDGFR and associated signals will be characterized as well. These results will be compared to findings obtained for tumor biopsy analysis to describe the correlation of these findings.

Bone marrow specimens should be obtained from the posterior iliac crest when possible. Aspiration should precede trephine biopsy collection. A total of 2-3 ml of marrow will be aspirated into a heparainzed syringe with no heparin remaining in the syringe at the time of collection. The

aspirate should be preserved in 0.5 ml aliquots stored in cyropresevation (Nunc) vials and will be stored in the laboratory of Dr. W. Douglas Figg until processing. The samples should be immediately frozen and stored at – 80 C until thawed for analysis. Biopsy material should be collected by a representative of Dr. Figg's laboratory. Garreth Peters (pager 104-4587-7 or 301-402-3622) should be notified for sample collection.

Trephine biopsies (bone marrow core biopsies) should be immediately preserved in OCT, barcoded, and stored in liquid nitrogen until the time of utilization.

Bone marrow biopsies will be lysed and printed on protein lysate arrays and analyzed as described below (section 7.2).

7.1.3 Leukocyte analysis

In addition to either direct tumor or bone marrow analysis, leukocytes will be isolated by buffy coat and lysed for correlative analysis of the Ras/Raf pathway elements, angiogenic markers, and RTK makers using the same protein microarrays proposed above as a surrogate to the bone marrow and tumor analysis. Approximately 10 ml of blood will be drawn using a CPT collection tube (Becton Dickinson). The blood will be centrifuged and the buffy coat layer of white blood cells will be removed, washed in cold PBS and pelleted by centrifugation. The DNA will be processed using a column purification technique (Qiagen), or the buffy coat pellet will be frozen for future DNA extraction.

7.1.3 Archival tissue

Archival tissue blocks will be collected for genomic DNA extraction for B-Raf mutational analysis in the laboratory of Dr. Paul Meltzer. This process will be streamlined using screening methods described in section 7.6.

7.2 **Pharmacoproteomic analysis**

7.2.1 The primary targets of pharmacoproteomic modulation to be measured were defined based upon the preclinical signaling data. They are listed below

Survival/Proliferation		**Angiogenesis**		**Apoptosis**		**Other**
AKT	p-AKT	eNOS	p-eNOS	PARP	Cleaved-PARP	PSA
ERK 1 / 2	p-ERK 1 / 2	PDGFRβ	p-PDGFRβ	Caspase 9	Cleaved Caspase-9	
Cyclin D1		CD31		Caspase 3	Cleaved Caspase-3	

7.2.2 Should additional tissue or TLA arrays be available, the following targets will be characterized. These will include but are not limited to the following:

Survival/Proliferation		**Angiogenesis**		**Apoptosis**		**Other**
mTOR	p-mTOR	α-smooth muscle actin		p53		PSMA
EGFR	p-EGFR	Pyk2	p-Pyk2	GSK3ß	p-GSK3ß	Cytokeratin-1
Src	p-Src	VEGFR-2		p-bad		
NFkB	p-NFkB			p-Bcl-2		
STAT1	p-STAT1					
TGFa						
p38	Phospho-p38					
Jak1	pJak1					
IkB	p-IkB					
IGFR						

7.2.3. Provided that the following antibodies be validated for usage and adequate tissue or TLA arrays be available, the following additional targets will be characterized:

Survival/Proliferation		**Angiogenesis**	**Apoptosis**
PCNA		αvβ3 complex	Bad
Fos		CD34	Annexin V
Raf		VEGFR-1	Bcl-2
Ras		bFGF	
CREB		vWF	
Rho		Factor VIII	
Jun	pJun		
pIkB			

Any unused tissue collected during the study will be saved for future studies, including assessment of the differences in gene expression/protein profiles in pre- and post-treatment using cDNA and protein microarray analyses in addition to other genomic and proteomic technologies. Consent for such procedures will be procured at the time of study enrollment.

7.4 Imaging studies

Non-invasive imaging has become a standard tool in the clinic to monitor primary and metastatic solid tumors. CT scan and bone scan will be performed on all patients at study entry and after every 2 cycles to assess clinical response to BAY 43-9006.

7.5 Assessment of the pharmacokinetic parameters of BAY 43-9006

Blood samples will be obtained serially from prior to ingestion until 24 hours post-dose at the following time points: immediately pre-dose, and at 0.25, 0.50, 1, 2, 4, 6, 8, 12, and 24 hours after ingestion, and then at each clinic visit. In the event that inpatient admission is not possible, PKs may be done on an outpatient basis. When outpatient PKs are being collected an hour 12 PK will not be required. The Clinical Pharmacology Research Core of Dr. William D. Figg will coordinate rapid acquisition and evaluation of patient samples. The pharmacokinetic characteristics of BAY 43-9006 in patients with AIPC will be evaluated using the WinNonlin software (Pharsight, Mountain View, CA). The maximum concentration, time to maximum concentration, the area under the curve extrapolated to infinity, and the apparent terminal half-life will be calculated. Pharmacodynamic study will address any correlation in BAY 43-9006 concentration with disease response and/or toxicities.

7.6 B-Raf mutation analysis

It has recently been found that B-Raf is commonly mutated in human tumors. To determine if BAY 43-9006 preferentially inhibits tumors with mutant B-Raf, mutational analysis of B-Raf using the following paradigm:

Initial screening for B-Raf mutation will be executed by protein lysate array analysis. The most frequently observed mutation in B-Raf is the V599E [31] amino acid substitution. Dr. Meltzer's group has developed a monoclonal antibody specific to this mutation (unpublished data). Lysate arrays will be optimized with this monoclonal antibody and then will be used to assess expression of this BRAF mutation. . Detection of this protein product will prompt allele specific PCR analysis of this region of the tumor genome using a recut from the tumor biopsies obtained for the protocol. No germline analysis will be done. Additionally, once a cohort of responders has been identified, genomic DNA will be extracted from tissue blocks for allele specific PCR in the other commonly mutated region, T1769A.

7.7

Labeling, Storage, and Tracking

- All labels used must be freezer-proof.
- Barcode labels will be used in this study. A sample of the barcode will be provided to the processing laboratory by Dr. Figg's laboratory before use because some DNA processing laboratories have encountered scanning incompatibility due to inclusion of hidden digits in barcode labels.
- Label each collection tube with the following information:
  - unique sample I.D.
  - study I.D. (NCI protocol number, etc.)
- All data associated with samples is entered into the Clinical Pharmacology Program's "Patient Sample Database Management System" (PSDMS) - Labrador. This is a secure program that can only be accessed by authorized users in Dr. Figg's lab. PSDMS creates a unique barcode ID for every sample and sample box which cannot be traced back to patients without PSDMS access

- The data recorded for each sample includes the patient ID, name, trial name/protocol number, time drawn, cycle time point, dose, material type, as well as box and freezer location. There are patient demographics that can be obtained to correlate with the samples through the PSDMS system. For each sample, there are associated processing notes (i.e., delay in sample processing, storage conditions on the ward, etc.). Bar-coded samples are stored in bar-coded boxes in a locked freezer at either -20 or -85°C according to stability requirements. These freezers are located onsite in Dr. Figg's lab and offsite at NCI Frederick Central Repository Services (Fisher Bioservices) in Frederick, MD. Samples will be stored until requested by the researcher assigned to the protocol (however, those requests must come from a member of Dr. Figg's laboratory with PSDMS access/clearance). All requests are monitored and tracked in the PSDMS system. All researchers are required to sign a form stating that the samples are only to be used for research purposes associated with this trial (as per the IRB approved protocol – that protocol is stored in the PSDMS system) and that any unused samples must be returned to Dr. Figg's laboratory.

- After collection, blood samples must be refrigerated (+ 4°C) or frozen (see chart below) at the site of collection and transported to the central reference laboratory or designated DNA processing laboratory **as soon as possible**.

Option	Storage Temperature at Treatment Site	Maximum Duration of Storage at Treatment Site	Transport Temperature	Delivery Time
1	+ 4°C (fridge)	24 hours	0 - 4°C (ice blocks)	24 hours
2	+ 4°C (fridge)	24 hours	Less than -20°C (dry ice)	24-72 hours

- **Samples must not be thawed and then re-frozen at any point.**

Protocol Completion/Sample Destruction

- Once primary research objectives for the protocol are achieved, intramural researchers can request access to remaining samples providing they have an IRB approved protocol and patient consent.

- Samples, and associated data, will be stored permanently unless the patient withdraws consent. If researchers have samples remaining once they have completed all studies associated with the protocol, they must be returned to Dr. Figg's laboratory.

The PI will report destroyed samples to the IRB if samples become unsalvageable because of environmental factors (ex. broken freezer or lack of dry ice in a shipping container) or if a patient withdraws consent. Samples will also be reported as lost if they are lost in transit between facilities or misplaced by a researcher. Dr. Figg's laboratory will report any freezer problems, lost samples or other problems associated with samples to the IRB, the NCI Clinical Director, and the office of the CCR, NCI.

8. STUDY CALENDAR

Schedules shown in the Study Calendar below are provided as an example and should be modified as appropriate (+/- 1-2 days to allow for clinic variations around federal holidays).

Baseline evaluations are to be conducted within 1 week prior to administration of protocol therapy. Scans and x-rays must be done 4 weeks prior to the start of therapy. In the event that the patient's condition is deteriorating, laboratory evaluations should be repeated within 48 hours prior to initiation of the next cycle of therapy.

	Pre-Study	C1D1	C1D2-28	C2D1	C3D1	C4D1	C5D1	C6D1	C7D1	Q4 weeks	Q12 weeks	Off Study[c]
BAY 43-9006		X	X	X	X	X	X	X	X	X	X	
Informed consent	X											
Demographics	X											
Medical history	X	X		X	X	X	X	X	X	X	X	X
Concurrent Meds	X	X	X	X	X	X	X	X	X	X	X	X
Physical exam	X	X		X	X	X	X	X	X	X	X	X
Vital signs	X	X		X	X	X	X	X	X	X	X	X
Height	X											
Weight	X	X		X	X	X	X	X	X	X	X	X
Performance status	X	X		X	X	X	X	X	X	X	X	X
CBC w/diff, plts, PT, PTT	X	X		X	X	X	X	X	X	X	X	X
Serum chemistry[b]	X	X		X	X	X	X	X	X	X	X	X
PSA	X	X		X	X	X	X	X	X	X	X	X
EKG (as indicated)	X											
Adverse event evaluation		X		X	X	X	X	X	X	X	X	X
Tumor measurements	X				X		X		X		X	
Radiological Evaluation	X				X		X		X	X	X	
Tumor Biopsy OR Bone Marrow Biopsy and Aspirate	X			X								
Other correlative studies		X		X								

a: BAY 43-9006: Dose as assigned; route/schedule.
b: Albumin, alkaline phosphatase, total bilirubin, bicarbonate, BUN, calcium, chloride, creatinine, glucose, LDH, phosphorus, potassium, total protein, SGOT(AST), SGPT(ALT), sodium, amylase, lipase
c: Off-study evaluation. Two consecutive measurements taken 4 weeks apart must be used to document progressive disease if the patient is removed from study for this reason.

9. MEASUREMENT OF EFFECT

For the purposes of this study, patients should be reevaluated for response every 8 weeks for the first 6 months of the study. After 6 months, CT and bone scans should be obtained at least every 3 months to monitor disease status while on BAY 43-9006. In addition to a baseline scan, confirmatory scans should also be 4 weeks following initial documentation of objective response.

9.1 Definitions

Response and progression will be evaluated in this study using the new international criteria proposed by the Response Evaluation Criteria in Solid Tumors (RECIST) Committee [*JNCI* 92(3):205-216, 2000]. Changes in only the largest diameter (unidimensional measurement) of the tumor lesions are used in the RECIST criteria. Note: Lesions are either measurable or non-measurable using the criteria provided below. The term "evaluable" in reference to measurability will not be used because it does not provide additional meaning or accuracy.

9.1.1 Measurable disease

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

9.1.2 Non-measurable disease

All other lesions (or sites of disease), including small lesions (longest diameter <20 mm with conventional techniques or <10 mm using spiral CT scan), are considered non-measurable disease. Bone lesions, leptomeningeal disease, ascites, pleural/pericardial effusions, lymphangitis cutis/pulmonis, inflammatory breast disease, abdominal masses (not followed by CT or MRI), and cystic lesions are all non-measurable.

9.1.3 Target lesions

All measurable lesions up to a maximum of five lesions per organ and 10 lesions in total, representative of all involved organs, should be identified as **target lesions** and recorded and measured at baseline. Target lesions should be selected on the basis of their size (lesions with the longest diameter) 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.

9.1.4 Non-target lesions

All other lesions (or sites of disease) should be identified as **non-target lesions** and should also be recorded at baseline. Non-target lesions include measurable lesions that exceed the maximum numbers per organ or total of all involved organs as well as non-measurable lesions. Measurements of these lesions are not required, but the presence or absence of each should be noted throughout follow-up.

9.2 Guidelines for Evaluation of Measurable Disease

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

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

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

by clinical examination when both methods have been used to assess the antitumor effect of a treatment.

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

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

Conventional CT and MRI. 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. Head and neck tumors and those of extremities usually require specific protocols.

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

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

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.

9.3 Response Criteria

9.3.1 Evaluation of target lesions

Complete Response (CR):	Disappearance of all target 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
Progressive Disease (PD):	At least a 20% increase in the sum of the LD of target lesions, taking as reference the smallest sum LD recorded since the treatment started 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

9.3.2 Evaluation of non-target lesions

Complete Response (CR):	Disappearance of all non-target lesions and normalization of tumor marker level
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

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

Note: If tumor markers are initially above the upper normal limit, they must normalize for a patient to be considered in complete clinical response.

9.3.3 **Evaluation of best overall response**

The best overall response is the best response recorded from the start of the treatment until disease progression/recurrence (taking as reference for progressive disease the smallest measurements recorded since the treatment started). The patient's best response assignment will depend on the achievement of both measurement and confirmation criteria (see section 9.3.1).

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

Note:

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

- In some circumstances, it may be difficult to distinguish residual disease from normal tissue. When the evaluation of complete response depends on this determination, it is recommended that the residual lesion be investigated (fine needle aspirate/biopsy) before confirming the complete response status.

9.4 Confirmatory Measurement/Duration of Response

9.4.1 Confirmation

To be assigned a status of PR or CR, changes in tumor measurements must be confirmed by repeat assessments that should be 4 weeks after the criteria for response are first met. In the case of SD, follow-up measurements must have met the SD criteria at least once after study entry at a minimum interval 8 weeks (see section 9.3.3).

9.4.2 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.

9.4.3 **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.

9.5 **Biochemical Response Criteria** –PSA measurements (PSA Consensus Criteria) (Bubley et al. 1999)

9.5.1 PSA Decline of ≥50%

A decline of PSA of at least 50% (confirmed by a second value at least 4 weeks after the first) with no other evidence of disease progression.

9.5.2. PSA Progression (all progression dates require to be confirmed by a second value after the first no sooner than 4 weeks after the initial measurement)

A 50% increase in PSA over nadir (confirmed by a second reading four weeks later) in patients whose PSA has fallen by at least 50%. PSA increase must be at least 5ng/ml.

9.5.2.1 A 25% increase in PSA over nadir (confirmed by a second reading) in patients whose PSA has not fallen by at least 50%. PSA increase must be at least 5ng/ml.

9.5.2.2 A 25% increase in PSA over baseline in patients whose PSA has not decreased. PSA increase must be at least 5ng/ml.

9.5.3 Time to PSA Progression

The time between the first day of treatment to the day of PSA progression as described in 9.5.2. the day of progression is defined as the first study day when the PSA level meets progression criteria (not the day of verification).

9.5.4 Per amendment D, Biochemical Response Criteria will continue to be measured as a primary endpoint, however patients may continue to receive BAY 43-9006 as long as they do not have clear evidence of radiographic or clinical progression.

9.6 **Progression-Free Survival**

The primary objective of this study is to determine whether BAY 43-9006, when used to treat metastatic androgen independent prostate cancer (AIPC), is associated with ≥50% of patients progression free at 4 months by PSA and clinical criteria. Patient monitoring studies and definitions of response are defined above. Please refer to the statistics section for further details.

Per Amendment D, a secondary objective of this study is to determine whether BAY 43-9006, when used to treat metastatic AIPC, is associated with ≥50% of patients progression free at 4 months by clinical and radiographic criteria. The 22 patients treated on the first stage of this protocol will be retrospectively evaluated with respect to this secondary endpoint. Please refer to the statistics section for further details.

9.7 **Response Review- N/A**

10. REGULATORY AND REPORTING REQUIREMENTS

Expedited adverse event (AE) reporting for this study is via AdEERS (Adverse Event Expedited Reporting System), accessed via the secure CTEP web site https://webapps.ctep.nci.nih.gov/openapps/plsql/gadeers_main$.startup). The reporting procedures to be followed are presented in the "NCI Guidelines: Expedited Adverse Event Reporting Requirements for NCI Investigational Agents" which can be downloaded from the *CTEP web site* (http://ctep.cancer.gov/reporting/adeers.html).

The descriptions and grading scales found in the revised NCI Common Terminology Criteria for Adverse

Events (CTCAE) version 3.0 will be utilized for adverse event reporting. All appropriate treatment areas should have access to a copy of the CTCAE version 3.0. A list of adverse events that have occurred or might occur (Reported Adverse Events and Potential Risks) can be found in Section 6 (Pharmaceutical Information). *A copy of the CTCAE version 3.0 can be downloaded from the CTEP web site (http://ctep.cancer.gov/reporting/ctc.html).*

10.1 Expedited Adverse Event Reporting

(AE; formerly known as Adverse Drug Reaction)

Expedited reports are submitted to CTEP via the secure AdEERS application accessed via the CTEP web site (https://webapps.ctep.nci.nih.gov/openapps/plsql/gadeers_main$.startup) and to the NCI IRB. Those AEs that do not require expedited reporting must be reported in routine (CDUS) study data submissions. AEs reported through AdEERS must **also** be reported in routine study data submissions.

10.1.1 Expedited Reporting Guidelines

Phase 2 and 3 Trials Utilizing an Agent under a CTEP IND: AdEERS Reporting Requirements for Adverse Events That Occur Within 30 Days[1] of the Last Dose of the Investigational Agent

	Grade 1	Grade 2	Grade 2	Grade 3		Grade 3		Grades 4 & 5[2]	Grades 4 & 5[2]
				Unexpected		Expected			
	Unexpected and Expected	Unexpected	Expected	with Hospitalization	without Hospitalization	with Hospitalization	without Hospitalization	Unexpected	Expected
Unrelated Unlikely	Not Required	Not Required	Not Required	10 Calendar Days	Not Required	10 Calendar Days	Not Required	10 Calendar Days	10 Calendar Days
Possible Probable Definite	Not Required	10 Calendar Days	Not Required	10 Calendar Days	10 Calendar Days	10 Calendar Days	Not Required	24-Hour; 5 Calendar Days	10 Calendar Days

[1] **Adverse events with attribution of possible, probable, or definite that occur greater than 30 days after the last dose of treatment with an agent under a CTEP IND require reporting as follows:**

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

- Grade 4 and Grade 5 unexpected events

AdEERS 10 calendar day report:

- Grade 3 unexpected events with hospitalization or prolongation of hospitalization
- Grade 5 expected events

[2] Although an AdEERS 24-hour notification is not required for death clearly related to progressive disease, a full report is required as outlined in the table.

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Note: All deaths on study must be reported using expedited reporting regardless of causality. Attribution to treatment or other cause should be provided.

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

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

expedited adverse event reporting exclusions.

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

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

10.1.2 Expedited Adverse Event Reporting Exclusions- N/A

10.1.3 Secondary AML/MDS

Investigators are required to report cases of secondary AML/MDS occurring on or following treatment on NCI-sponsored chemotherapy protocols using the NCI/CTEP Secondary AML/MDS Report Form. *This form can be downloaded from the CTEP web site (http://ctep.cancer.gov/reporting/index.html).* Second malignancies and non-AML/MDS secondary malignancies (*e.g.*, endometrial cancer in a breast cancer patient receiving tamoxifen) should NOT be reported via AdEERS but should be submitted as part of the study results via routine CDUS reporting.

10.2 Data Reporting

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

10.3 CTEP Multicenter Guidelines- N/A

10.4 Cooperative Research and Development Agreement (CRADA)/Clinical Trials Agreement (CTA)

The agent(s) supplied by CTEP, DCTD, NCI used in this protocol is/are provided to the NCI under a Collaborative Agreement (CRADA, CTA) between the Pharmaceutical Company(ies) (hereinafter referred to as ACollaborator(s)@) and the NCI Division of Cancer Treatment and Diagnosis. Therefore, the following obligations/guidelines, in addition to the provisions in the AIntellectual Property Option to Collaborator@ contained within the terms of award, apply to the use of the Agent(s) in this study:

1. Agent(s) may not be used for any purpose outside the scope of this protocol, nor can Agent(s) be transferred or licensed to any party not participating in the clinical study. Collaborator(s) data for Agent(s) are confidential and proprietary to Collaborator(s) and shall be maintained as such by the investigators. The protocol documents for studies utilizing investigational Agents contain confidential information and should not be shared or distributed without the permission of the NCI. If a copy of this protocol is requested by a patient or patient's family member participating on the study, the individual should sign a confidentiality agreement. A suitable model agreement can be downloaded from: http://ctep.cancer.gov
2. For a clinical protocol where there is an investigational Agent used in combination with (an)other investigational Agent(s), each the subject of different collaborative agreements , the access to and use of data by each Collaborator shall be as follows (data pertaining to such combination use shall hereinafter be referred to as "Multi-Party Data.@):
    a. NCI must provide all Collaborators with prior written notice regarding the existence and nature of any agreements governing their collaboration with NIH, the design of the proposed combination protocol, and the existence of any obligations that would tend to restrict NCI's participation in the proposed combination protocol.

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

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

3. Clinical Trial Data and Results and Raw Data developed under a collaborative agreement will be made available exclusively to Collaborator(s), the NCI, and the FDA, as appropriate. All data made available will comply with HIPAA regulations

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

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

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

Regulatory Affairs Branch, CTEP, DCTD, NCI
Executive Plaza North, Suite 7111
Bethesda, Maryland 20892
FAX 301-402-1584
Email: anshers@ctep.nci.nih.gov

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

11. STATISTICAL CONSIDERATIONS

11.1 Study Design/Endpoints

The study was originally designed to have a two-stage design with 22 and then up to a total of 46 patients evaluating progression using PSA criteria. The results from the first stage were inadequate to justify accrual to the second stage. However, per amendment D, waiver of the stopping rule is requested to allow for accrual to the second stage.

The primary objective of this study is to determine whether BAY 43-9006, when used to treat metastatic AIPC, is associated with having 50% of patients progression free at 4 months by clinical, radiographic and PSA criteria. The secondary objective of this study is to determine the time to progression measured by

clinical and radiographic criteria. If this is found to be the case, then this agent will be considered potentially useful when combined with other anticancer agents to be tested in later trials. Data from previous trials of NCI patients with similar eligibility requirements demonstrated a 2.1 month median progression free survival when using thalidomide (n=63) [32]. Median progression free survival on ketoconazole alone is expected to be 4 months based on previous trials[33]. Ketoconazole is recognized as a standard therapy alterative to cytotoxic chemotherapy in the setting of failure of primary hormonal therapy with GnRH agonists and anti-androgen therapy. The Eastern Cooperative Oncology Group is currently sponsoring a phase III clinical trial comparing ketoconazole to the combination of estramustine and docetaxel. Median progression free survival on ketoconazole alone is expected to be 4 months based on previous trials [34, 35]. Based on these results, it would be useful to demonstrate whether BAY 43-9006 is able to induce progression free survival in 50% of patients at the 4 month (approximately day 113 evaluation) time point.

The study will be conducted as a two-stage optimal design (Simon 1989)[29]. Using alpha=0.10 and beta=0.10 as acceptable error probabilities, the trial will target 50% as the desirable proportion of patients who are still without progression by clinical, radiographic and PSA criteria at the 4th monthly evaluation (p1=0.50; see section 9.5.1 for details), and will be considered inadequate if only a fraction consistent with 30% are without progression by the same evaluation time (p0=0.30).

> Initially, 22 patients will be enrolled and evaluated for progression. The enrollment will be temporarily halted after the 22nd patient unless we know that 8 patients have passed the 4 month point without progression. If 8 or more of the first 22 patients enrolled on the trial have not progressed as defined in Section 9.3 and 9.4 at the 4th evaluation (day 113, approximately), then enrollment will continue until 46 patients have been enrolled. If, among the first 22 patients enrolled, 7 or fewer are able to be progression free at the 4 month evaluation, then the original design specified that no further patients will be enrolled once such a determination has been made. If 18 or more of the total cohort of 46 patients have been found to be progression free at 4 months, then this will indicate an adequate progression free probability to be worthy of further consideration in future trials. On the other hand, if 8-17 of 46 are progression free at 4 months, this will be considered insufficient. Under the null hypothesis (30% progression free at 4 months), the probability of being able to stop accrual after 22 patients have been evaluated at 4 months is 67%. However, under amendment D, the stopping rule will be waived due to potential benefit identified in patients during the first stage, despite failure to meet criteria established for continuing accrual. The total set of 46 patients will be evaluated with respect to progression free survival, using clinical, radiographic and PSA criteria to primarily focus on 4 month progression free survival. In addition, progression free survival evaluated by clinical and radiographic measures only will be determined on all evaluable patients from whom this may be determined.

Given the difficulties in interpreting progression free rates in our patients compared to historical data, the response rate will be included as a secondary clinical endpoint.

> In addition to evaluation of the proportion progression free at 4 months, the progression free survival will also be analyzed via a Kaplan-Meier curve. This will be done for both progression free survival evaluated by clinical, radiographic and PSA criteria, as well as secondarily based only on clinical and radiographic criteria. This latter, secondary, evaluation will be based on all patients who are able to be assessed by these criteria in either the first (retrospectively) or second stage of accrual.

11.2 **Sample Size/Accrual Rate**

Based on previous efforts in recruiting patients with this disease onto trials at the NCI, it is anticipated that 30 patients per year may be able to enroll onto this protocol. Thus, it is expected that accrual to this trial can be completed in approximately 1.5 years if all 46 patients are to be enrolled

11.3 **Stratification Factors**

There are no stratification factors

11.4 **Analysis of Secondary Endpoints**

Proteomic, PSA, pharmacokinetic, and molecular endpoints will be evaluated on the protocol in all available enrolled patients. These will all be considered exploratory analyses, and will not have their statistical results adjusted for multiple comparisons. However, all interesting findings will be carefully interpreted as hypothesis generating.

11.5 **Reporting and Exclusions**

11.5.1 **Evaluation of toxicity.** All patients will be evaluable for toxicity from the time of their first treatment with BAY 43-9006

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

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

12. HUMAN SUBJECTS PROTECTIONS

12.1 Rationale for subject selection

Subjects treated on this study, will be individuals with metastatic prostate cancer, which has recurred (or persisted) after appropriate standard treatment. Individuals of any race or ethnic group will be eligible for this study. Eligibility assessment will be based solely on the patient's medical status. Recruitment of patients onto this study, will be through standard CCR mechanisms. No special recruitment efforts will be conducted.

12.2 Evaluation of benefits and risks/discomforts

The potential benefit to a patient that goes onto study, is a reduction in the bulk of their tumor and improvement in their bony lesions, which may or may not have favorable impact on symptoms and/or survival. Potential risks include the possible occurrence of any of a range of side effects, that are listed in the Consent Document (Please refer to http://ctep.cancer.gov/reporting/adeers.html to review the up-to-date expected adverse event list for BAY 43-9006). The procedure for protecting against or minimizing risks, will be to medically evaluate patients on a regular basis as described in Section 9.

12.3 Risks to patients in relation to anticipated benefits

12.3.1 For patients with androgen-independent prostate cancer, median survival is in the range of 12-18 months. Potential risks include the possible occurrence of any of a range of side effects listed in section 6. Please refer to http://ctep.cancer.gov/reporting/adeers.html to review the up-to-date expected adverse event list for BAY 43-9006.

12.3.2 Risk of serial biopsies: All care will be taken to minimize risks that may be incurred by tumor sampling. However, there are procedure-related risks (such as bleeding, infection and visceral injury) that will be explained fully during informed consent. If patients suffer any physical injury as a result of the biopsies, immediate medical treatment is available at the NIH's Clinical Center in Bethesda, Maryland. Although no compensation is available, any injury will be fully

evaluated and treated in keeping with the benefits or care to which patients are entitled under applicable regulations.

12.4 Consent process

Patients will meet with an associate or principal investigator on the trial in the Prostate Cancer Clinic, during the initial evaluation for this study. During that meeting, the investigator will inform patients of the purpose, alternatives, treatment plan, research objectives and follow-up of this trial. The investigator will then provide a copy of the IRB-approved informed consent document that is included in this protocol. The patient will be allowed to take as much time as he wishes, in deciding whether or not he wishes to participate. If a prolonged period of time expires during the decision making process (several weeks, as an example), it may be necessary to reassess the patient for protocol eligibility. A copy of the signed informed consent will be placed in the patient's medical record and the original held in the Protocol Office.

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

12.6 Patient Records and Quality Assurance

Quality assurance complete records must be maintained on each patient treated on the protocol. These records should include primary documentation (e.g.: laboratory report slips, X-ray reports, scan reports, pathology reports, physicians notes, etc.) which confirm that:

12.6.1 The patient met all eligibility criteria

12.6.2 Signed informed consent was obtained prior to treatment

12.6.3 Treatment was given according to protocol (dated notes about doses given, complications, and clinical outcomes).

12.6.4 Toxicity was assessed according to protocol (laboratory report slips, etc.)

12.6.5 Response was assessed according to protocol (X-ray, scan, lab reports, date noted on clinical assessment, as appropriate).

12.6.6 Drug Accountability

The unused BAY 43-9006 (partial bottles, empty bottles, and full bottles) will be returned for drug accountability at each clinic visit. An oral study agent case report form will be used to document drug accountability for each patient on this study. Unused agent, which is not returned to the patient for the next dose cycle, will be disposed of according to the Procedure of Disposal of Returned Oral agents.

12.6.7 Patients will use a diary to document daily drug intake and adverse events.

13. Data and Safety Monitoring Plan

A complete CDUS report will be submitted to CTEP on a quarterly basis. Response data will be on a quarterly basis as well. All adverse drug reactions will be reported to the NCI Institutional Review Board (IRB) within 10 working days of the date of occurrence. Safety information from patients enrolled onto the study will be gathered and reported. Any unusual toxicity will be explored for possible causative mechanisms. The safety of the repetitive biopsy procedure will also be determined and reported.

13.1 Any unanticipated or unknown treatment- or drug-related toxicity(ies) and life-threatening and lethal toxicity(ies) will be reported according to the DCT's Guidelines for Reporting Adverse Drug Reactions to the Investigational Drug Branch (301-230-2330; Fax 301-230-0159) within 24 hours, with a copy sent to the NCI-IRB. All fatal events (NCI/CTEP grade V toxicity) will require a written report from the

Principal or Associate Investigator will follow within 10 working days. A written report, within 10 working days, should be sent on CTEP ADR forms to:

Investigational Drug Branch
PO Box 30012
Bethesda, MD 20824

Clinical Associates and/or senior staff should notify the Principal Investigator at 301-435-8183 (Dr. Dahut), Bldg.10, Rm 12N226 of the occurrence of such toxicity.

13.2 Adverse events listed in section 6 do not require ADEERS reporting except as otherwise mandated by the guidelines (e.g., grade 3 with hospitalization, grade 4 or 5). The occurrence of any new toxicity (not in the above list), regardless of grade, and all life threatening events (NCI/CTEP grade IV toxicity) will be reported to the Drug Monitor, Investigational Drug Branch, CTEP, and the NCI IRB within 24 hours. An ADR form may be required. The Principal Investigator, Study Coordinator, or an Associate Investigator will be responsible for completing the ADR form and for notifying the NCI's Institutional Review Board.

A summary of the ongoing study will be submitted to the NCI's Institutional Review Board at 12 month intervals and a final report will be sent within six months of study completion at the request of the Institutional Review Board using the CTEP study summary form. The status reported will be submitted and presented at upcoming NCI meetings as requested

References

1. Jemal, A., et al., *Cancer statistics, 2003.* CA Cancer J Clin, 2003. **53**(1): p. 5-26.
2. Rini, B.I. and E.J. Small, *Prostate cancer update.* Curr Opin Oncol, 2002. **14**(3): p. 286-91.
3. Goktas, S. and E.D. Crawford, *Optimal hormonal therapy for advanced prostatic carcinoma.* Semin Oncol, 1999. **26**(2): p. 162-73.
4. Klotz, L., *Hormone therapy for patients with prostate carcinoma.* Cancer, 2000. **88**(12 Suppl): p. 3009-14.
5. Beedassy, A. and G. Cardi, *Chemotherapy in advanced prostate cancer.* Semin Oncol, 1999. **26**(4): p. 428-38.
6. *Investigator's Brochure, versions 3 and 4.1*. ed. A. Shah, C. Lathia, and K. Heininger. 2003, Bayer Health Care: West Haven, CT.
7. Campbell, S.L., et al., *Increasing complexity of Ras signaling.* Oncogene, 1998. **17**(11 Reviews): p. 1395-413.
8. Chong, H., H.G. Vikis, and K.L. Guan, *Mechanisms of regulating the Raf kinase family.* Cell Signal, 2003. **15**(5): p. 463-9.
9. Cohen, Y., et al., *BRAF mutation in papillary thyroid carcinoma.* J Natl Cancer Inst, 2003. **95**(8): p. 625-7.
10. Davies, H., et al., *Mutations of the BRAF gene in human cancer.* Nature, 2002. **417**(6892): p. 949-54.
11. Mercer, K.E. and C.A. Pritchard, *Raf proteins and cancer: B-Raf is identified as a mutational target.* Biochim Biophys Acta, 2003. **1653**(1): p. 25-40.
12. Pollock, P.M. and P.S. Meltzer, *A genome-based strategy uncovers frequent BRAF mutations in melanoma.* Cancer Cell, 2002. **2**(1): p. 5-7.
13. Sawyers, C.L., *Finding the next Gleevec: FLT3 targeted kinase inhibitor therapy for acute myeloid leukemia.* Cancer Cell, 2002. **1**(5): p. 413-5.
14. Singer, G., et al., *Mutations in BRAF and KRAS characterize the development of low-grade ovarian serous carcinoma.* J Natl Cancer Inst, 2003. **95**(6): p. 484-6.
15. Tannapfel, A., et al., *Mutations of the BRAF gene in cholangiocarcinoma but not in hepatocellular carcinoma.* Gut, 2003. **52**(5): p. 706-12.
16. Bergers, G., et al., *Benefits of targeting both pericytes and endothelial cells in the tumor vasculature with kinase inhibitors.* J Clin Invest, 2003. **111**(9): p. 1287-95.
17. Ferrara, N., *VEGF and the quest for tumour angiogenesis factors.* Nat Rev Cancer, 2002. **2**(10): p. 795-803.
18. Apperley, J.F., et al., *Response to imatinib mesylate in patients with chronic myeloproliferative diseases with rearrangements of the platelet-derived growth factor receptor beta.* N Engl J Med, 2002. **347**(7): p. 481-7.
19. Heinrich, M.C., et al., *Inhibition of KIT tyrosine kinase activity: a novel molecular approach to the treatment of KIT-positive malignancies.* J Clin Oncol, 2002. **20**(6): p. 1692-703.
20. Alavi, A., et al., *Role of Raf in vascular protection from distinct apoptotic stimuli.* Science, 2003. **301**(5629): p. 94-6.
21. Bushman, E.C., R.N. Nayak, and W. Bushman, *Immunohistochemical staining of ras p21: staining in benign and malignant prostate tissue.* J Urol, 1995. **153**(1): p. 233-7.
22. Agnantis, N.J., et al., *Comparative immunohistochemical study of ras-p21 oncoprotein in adenomatous hyperplasia and adenocarcinoma of the prostate gland.* Anticancer Res, 1994. **14**(5B): p. 2135-40.
23. Bakin, R.E., et al., *Constitutive activation of the Ras/mitogen-activated protein kinase signaling pathway promotes androgen hypersensitivity in LNCaP prostate cancer cells.* Cancer Res, 2003. **63**(8): p. 1981-9.
24. Bakin, R.E., et al., *Attenuation of Ras signaling restores androgen sensitivity to hormone-refractory C4-2 prostate cancer cells.* Cancer Res, 2003. **63**(8): p. 1975-80.
25. Fu, Z., et al., *Effects of raf kinase inhibitor protein expression on suppression of prostate cancer metastasis.* J Natl Cancer Inst, 2003. **95**(12): p. 878-89.
26. Qi, L., et al., *Migration and invasion of human prostate cancer cells is related to expression of VEGF and its receptors.* Anticancer Res, 2003. **23**(5A): p. 3917-22.
27. Wu, J., K. Haugk, and S.R. Plymate, *Activation of pro-apoptotic p38-MAPK pathway in the prostate cancer cell line M12 expressing a truncated IGF-IR.* Horm Metab Res, 2003. **35**(11-12):

p. 751-7.
28. Nicholson, B. and D. Theodorescu, *Angiogenesis and prostate cancer tumor growth.* J Cell Biochem, 2004. **91**(1): p. 125-50.
29. Simon, R., *Optimal two-stage designs for phase II clinical trials.* Control Clin Trials, 1989. **10**(1): p. 1-10.
30. Brown, R.S., et al., *The comparative values of bone marrow aspirate and trephine for obtaining bone scan-targeted metastases from hormone-refractory prostate cancer.* Prostate Cancer Prostatic Dis, 2002. **5**(2): p. 144-51.
31. Pollock, P.M., et al., *High frequency of BRAF mutations in nevi.* Nat Genet, 2003. **33**(1): p. 19-20.
32. Figg, W.D., et al., *A randomized phase II trial of thalidomide, an angiogenesis inhibitor, in patients with androgen-independent prostate cancer.* Clin Cancer Res, 2001. **7**(7): p. 1888-93.
33. Figg, W.D., et al., *A randomized, phase II trial of ketoconazole plus alendronate versus ketoconazole alone in patients with androgen independent prostate cancer and bone metastases.* J Urol, 2005. **173**(3): p. 790-6.
34. Walczak, J.R. and M.A. Carducci, *Phase 3 randomized trial evaluating second-line hormonal therapy versus docetaxel-estramustine combination chemotherapy on progression-free survival in asymptomatic patients with a rising prostate-specific antigen level after hormonal therapy for prostate cancer: an Eastern Cooperative Oncology Group (E1899), Intergroup/Clinical Trials Support Unit study.* Urology, 2003. **62 Suppl 1**: p. 141-6.
35. Small, E.J., et al., *Ketoconazole retains activity in advanced prostate cancer patients with progression despite flutamide withdrawal.* J Urol, 1997. **157**(4): p. 1204-7.

APPENDIX A

Performance Status Criteria

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

Appendix B: NCI/FDA Proteomics Program Standard Operating Procedure for Tissue Core Collection- Needle Biopsy – Cryopreservation in OCT

Principle:

Core needle biopsies are used to sample tissue from a specific, defined location. These biopsies may consist of normal, pre-malignant and malignant tissue due to the multi-level tissue sample that is obtained. This type of sample is ideal for studying the micro-tumor environment.

Rapid freezing of the sample is required to prevent degradation of the proteins or RNA. Optimal Cutting Temperature (OCT) compound is an alcohol polymer that is liquid at room temperature and a solid at –20^{o}C. This polymer is used to cryo-protect the tissue and provide a medium for cryo-sectioning.

Materials:

Cryomolds (Sakura Finetek Ca.t # 4728)

OCT (Sakura Finetek Cat. # 4583)

Dry ice

Ultra cold freezer (-70^{0} to -80^{0}C)

Needle: 16 or 18 gauge

Permanent marker

Sterile forceps

Sterile Glass slides

Aluminum foil or 50ml Falcon tubes

Procedure:

1. Prepare all supplies prior to the biopsy procedure to avoid delay once the specimen has been obtained.
2. Label the handle and the front surface of the cryomold with the sample or patient's identifying information.
3. Perform core needle biopsy.
4. Pick the core from the biopsy needle onto a sterile glass slide.
5. Fill cryomold about 1/3 full with OCT. Place the cryomold in dry ice to partially freeze the OCT. The OCT should be jelly-like, not completely frozen.
6. Carefully lift the core biopsy by both ends with sterile forceps. **Do not stretch the biopsy or it will break.**
7. Lay the biopsy as straight as possible in the OCT. Once the sample touches the OCT, you cannot reposition it or the sample will break apart.
8. Quickly add OCT on top of the biopsy, completely covering the sample.
9. Ensure the sample is level and freeze immediately in dry ice.
10. Store wrapped in aluminum foil or in a 50ml Falcon tube at –70^{o}C.

Note:

Do not lay the biopsy on frozen OCT and cover it with liquid OCT. The OCT will not fuse and will split into two sections when cutting the frozen tissue sections.

Frozen Section Slides

1. Frozen sections for proteomic analysis should be cut at 5-8um on plain, uncoated glass microscope slides.
2. The tissue section should be placed as close as possible to the center of the slide. Do not place the frozen section at the end of the slide.
3. Two tissue sections from the same biopsy may be placed on the same glass slide if space permits.
4. Do not allow the tissue section to air on the slide. Freeze immediately on dry ice or at –80^{o}C.

Shipping Slides or Frozen Tissue

1. Ship slides/tissue in OCT on dry ice Monday through Thursday only.
2. Tissue should be embedded in OCT prior to shipment. Refer to procedure steps 5-10.
3. Seal slide box in a plastic bag with dessicant (such as Drierite crystals). Seal tissue in a 50ml polypropylene Falcon tube, wrap in aluminum foil or place in a plastic bag.
4. Place dry ice on top of the plastic bag containing the slides/tissue.
5. Place any special instructions/inventory/shipping documents in the box.
6. Seal the box with tape.
7. Ship to:

Ginny Espina
National Cancer Institute
9000 Rockville Pike
Bldg 10 Room B1B53
Bethesda, MD 20892

301-435-7763

Appendix C: Cytochrome P450 3A4 Metabolized Drugs

Substrates

Acetaminophen	Dapsone	Levobupivicaine	Risperidone
Alfentanil	Dehydroepiandrostendione	Lidocaine	Ritonavir**
Alosetron	Delavirdine	Loratadine	Salmeterol
Alprazolam	Desmethyldiazepam	Losartan	Saquinavir
Amiodarone	Dexamethasone	Lovastatin	Sertindole
Amitriptyline (minor)	Dextromethorphan (minor)	Methadone	Sertraline
Amlodipine	Diazepam (minor)	Mibefradil	Sibutramine
Anastrozole	Digitoxin	Miconazole	Sildenafil citrate
Androsterone	Diltiazem	Midazolam	Simvastatin
Antipyrine	Disopyramide	Mifepristone	Sirolimus
Astemizole	Docetaxel	Mirtazapine	Sufentanil
Atorvastatin	Dofetilide (minor)	Montelukast	Tacrolimus
Benzphetamine	Dolasteron	Navelbine	Tamoxifen
Bepridil	Donepezil	Nefazodone	Temazepam
Bexarotene	Doxorubicin	Nelfinavir	Teniposide
Bromazepam	Doxycycline	Nevirapine	Terfenadine
Bromocriptine	Dronabinol	Nicardipine	Testosterone
Budesonide	Enalapril	Nifedipine	Tetrahydrocannabinol
Bupropion (weak)	Erythromycin	Niludipine	Theophylline
Buspirone	Estradiol	Nimodipine	Tiagabine
Busulfan	Ethenyl estradiol	Nisoldipine	Tolterodine
Caffeine	Ethosuximide	Nitrendipine	Toremifene
Cannabinoids	Etoposide	Omeprazole (sulfonation)	Trazodone
Carbamazepine	Exemestane	Ondasetron	Tertinoin
Cerivastatin	Felodipine	Oral contraceptives	Triazolam
Cevimeline	Fentanyl	Orphenadrine	Troglitazone
Chlorpromazine	Fexofenadine	Paclitaxel	Troleandomycin
Cimetidine	Finasteride	Pantoprazole	Venlafaxine
Cisapride	Fluoxetine	Pimozide	Verapamil
Citalopram	Flutamide	Pioglitazone	Vinblastine
Clarithromycin	Glyburide	Pravastatin	Vincristine
Clindamycin	Granisetron	Prednisone	Warfarin
Clomipramine	Halofantrine	Progesterone	Yohimbine
Clonazepam	Hydrocortisone	Proguanil	Zaleplon (minor)
Clozapine	Hydroxyarginine	Propafenone	Zatoestron
Cocaine	Ifosfamide	Quercetin	Zileuton
Codeine	Imipramine	Quetiapine	Ziprasidone
Cortisol	Indinavir	Quinidine	Zolpidem
Cortisone	Isradipine	Quinine	Zonisamide
Cyclobenzaprine	Itraconazole	Repaglinide	
Cyclophosphamide	Ketoconazole	Retinoic acid	
Cyclosporine	Lansoprazole (minor)	Rifampin	

Inducers

Carbamazepine	Nelfinavir	Primidone	Sulfadimidine
Dexamethasone	Nevirapine	Progesterone	Sulfinpyrazone
Ethosuximide	Oxcarbazepine	Rifabutin	Troglitazone
Glucocorticoids	Phenobarbital	Rifampin	
Griseofulvin	Phenylbutazone	Rofecoxib (minor)	
Nafcillin	Phenytoin	St. John's Wort	

Inhibitors

Amiodarone	Disulfiram	Mibefradil	Ranitidine
Anastrozole	Entacapone	Miconazole (moderate)	Ritonavir**
Azithromycin	Erythromycin	Nefazodone	Saquinavir
Cannabinoids	Ethenyl estradiol	Nelfinavir	Sertindole
Cimetidine	Fluconazole (weak)	Nevirapine	Sertraline
Clarithromycin	Fluoxetine	Norfloxacin	Troglitazone
Clotrimazole	Fluvoxamine	Norfluoxetine	Troleandomycin
Cyclosporine	Gestodene	Omeprazole (weak)	Valproic acid (weak)
Danazole	Grapefruit Juice	Oxiconazole	Verapamil
Delavirdine	Indinavir	Paroxetine (weak)	Zarfirlukast
Dexamethasone	Isoniazid	Propoxyphene	Zileuton
Dimethyldithiocarbamate	Itraconazole**	Quinidine	
Diltiazem	Ketoconazole**	Quinin	
Dirithromycin	Metronidazole	Quinupristin and dalfopristin	

** Contraindications
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