solid tumor panels. Using a time to event measure of efficacy, AZD2171 had intermediate and high levels of activity against 10 and 1 of 26 solid tumor xenografts evaluable for this measure, respectively. Intermediate activity was observed in 4 of 5 rhabdomyosarcoma, 3 of 3 Ewing sarcoma, and 2 of 3 Wilms tumor (WT) xenografts, with high level activity observed in 1 of 2 evaluable rhabdoid tumor (RT) xenografts. AZD2171 induced CR against 1 of 3 osteosarcoma (OS), 1 of 3 RT, and 1 of 3 WT xenografts, but had no effect on *in vivo* growth of any ALL xenografts. Kasumi-1, the only PPTP *in vitro* panel cell line with an $EC_{50} < 1\,\mu M$ (EC_{50} 0.175 μM) is known to have a gain-of-function $KIT^{Asn822Lys}$ mutation.
Conclusions: AZD2171 demonstrated broad activity against the PPTP's solid tumor panel. Antitumor activity was manifested primarily as tumor growth delay, although tumor regressions were observed in the OS, RT, and WT panels. Further preclinical evaluation of AZD2171 is warranted and will include studies of AZD2171 in combination with clinically relevant agents for selected xenografts in which activity was observed. The Pediatric Brain Tumor Consortium is planning clinical evaluations of AZD2171 in children.
Supported by NCI NO1CM42216.

103 POSTER

A phase II study of enzastaurin as second- or third-line treatment of non-small cell lung cancer (NSCLC)

Y. Oh[1], R. Herbst[1], H. Burris[2], A. Cleverly[3], M. Lahn[3], G. Bepler[4].
[1]*M.D. Anderson Cancer Center, Houston, TX, USA;* [2]*Sarah Cannon Research Institute, Nashville, TN, USA;* [3]*Eli Lilly and Company, Oncology, Indianapolis, IN, USA;* [4]*H. Lee Moffitt Cancer Center, Tampa, FL, USA*

Background: Enzastaurin, an oral serine/threonine kinase inhibitor, targets the PKC and PI3K/AKT pathways to inhibit tumor cell survival and proliferation and tumor-induced angiogenesis. PKC isoforms and PI3K/AKT are over-expressed and active in NSCLC and are associated with poor prognosis and treatment resistance. Based on preclinical and phase I trial data, this multicenter phase II trial was conducted to evaluate enzastaurin as second- and third-line treatment of NSCLC. The primary objective was to determine the rate of progression-free survival (PFS) at 6 months. Secondary objectives included safety, antitumor activity, and the rate of survival at 12 months.
Methods: Patients with advanced (stage IIIB) or metastatic (stage IV) NSCLC, who had failed at least 1 prior therapy, received 500 mg of oral enzastaurin (tablets), once daily after breakfast, every 28 days until disease progression or unacceptable toxicity occurred. All patients had to have failed prior platinum-based chemotherapy and had to be considered eligible for second- or third-line treatment.
Results: Fifty-three patients (55% male, 45% female; ECOG $\leqslant 2$), were enrolled. All patients had prior chemotherapy, including 28% with EGFR inhibitor treatment. At the time of interim analysis, 34 (64%) patients were alive. The median PFS time was 1.84 months (95% limits: 1.68–1.87 months) and the rate of PFS at 6 months was 10.4% (95% limits: 8.4%-10.9%). Eighteen patients (34%) had a best response of stable disease, while no patients had a partial or complete response. Of the 53 patients, 10 (19%) received therapy for $\geqslant 6$ cycles, 3 of whom are currently receiving enzastaurin beyond 9 cycles of treatment. The most common toxicity was fatigue (n = 21), noted within 1 week of starting treatment, but was not reported in patients with disease stabilization.
Conclusion: Although no objective tumor responses were observed in this study, 10.4% of the patients were progression-free at 6 months. Additional evaluations are ongoing to better understand the use of enzastaurin in the treatment of NSCLC, including studies to determine how to best combine enzastaurin with other agents active in NSCLC.

104 POSTER

Simultaneous blockade of VEGF and HGF receptors results in potent anti-angiogenic and anti-tumor effects

A. Joly. *Exelixis Inc., Drug Discovery, South San Francisco, USA*

Vascular endothelial growth factor (VEGF) and its tyrosine kinase receptors, VEGFR1 and VEGFR2, are expressed on the surface of vascular endothelial cells and play a central role in the promotion of tumor angiogenesis. Hepatocyte growth factor (HGF) and its tyrosine kinase receptor Met are overexpressed or activated in a wide variety of tumor types, promoting tumor growth. Additionally, HGF and VEGF interact synergistically to promote endothelial cell proliferation, tubule formation, and growth of new vessels in vivo. EXEL-7184 (XL184) is a small molecule tyrosine kinase inhibitor that targets the VEGF and HGF receptor tyrosine kinases.
In enzymatic assays, EXEL-7184 is a potent inhibitor of VEGFR2 and Met tyrosine kinase activities, with IC_{50} values in the sub-nanomolar and single digit nanomolar range, respectively. EXEL-7184 also exhibits potent activity in cell based assays, inhibiting VEGF-induced activation of ERK in endothelial cells, and HGF-induced activation of Met in tumor cells. EXEL-7184 also inhibits endothelial tubule formation stimulated by VEGF or by conditioned media derived from tumor cell lines, and HGF induced responses in tumor cells (e.g. invasion, chemotaxis). In pharmacodynamic assays, oral administration of EXEL-7184 resulted in dose-dependent and reversible inhibition of VEGFR2 in mouse lung, and of Met in xenograft tumors and in mouse liver. Following acute administration to xenograft tumor-bearing mice, EXEL-7184 caused rapid disruption of the tumor vasculature, and apoptosis of both tumor and endothelial cells. Administration of EXEL-7184 using both once-daily and less frequent oral dosing schedules resulted in significant efficacy in a range of solid tumor models, with substantial regression of large tumors. Immunohistochemical analysis at the end of efficacy studies demonstrated potent inhibitory effects on the tumor vasculature, and strong induction of tumor cell death.
In summary, EXEL-7184 is a potent inhibitor of VEGFR2 and Met, and exhibits potent anti-tumor and anti-angiogenic activity in preclinical models. A phase I clinical trial for EXEL-7184 is in progress.

105 POSTER

A prospective study of the cutaneous side effects of sorafenib, a novel multi-kinase inhibitor

J. Autier[1], B. Escudier[1], A. Spatz[1], B. Schwartz[2], C. Robert[1]. [1]*Institut Gustave Roussy, Villejuif, France;* [2]*Bayer Pharmaceuticals Corporation, West Haven, CT, USA*

Background: This study investigated the incidence, severity and management of cutaneous reactions to the oral multi-kinase inhibitor sorafenib (Nexavar®) (SOR) in advanced renal cell carcinoma (RCC) patients. Additional objectives were to determine the effect of SOR on nevi and on ERK phosphorylation in normal skin.
Materials and Methods: During a 16 month period from Nov. 2003 to Feb. 2005, all consecutive patients included in our center in the Phase III TARGETs RCC trial were enrolled in this dermatologic sub-study. Patients were randomized to receive either SOR 400 mg twice daily (bid) or placebo. Dermatologic examination was performed before treatment, every 3 weeks during the first four cycles, and every 4 weeks thereafter.
Results: Ninety-six patients with unresectable or metastatic RCC and failure of one systemic therapy were enrolled, and 85 received either SOR (n = 43) or placebo (n = 42). Thirty-nine patients (90.7%) on SOR, and three (7.1%) on placebo, had $\geqslant$ one cutaneous adverse event. Most cutaneous adverse events were mild or moderate; only two SOR patients had grade 3 toxicity (hand–foot skin reaction [HFSR]). No patient discontinued due to cutaneous side effects. HFSR was typically less severe and more localized than the form associated with traditional chemotherapies. HFSR (grade 3) led to dose reductions in two patients (50%), but resolved in 3–4 weeks without sequelae. After restoration of full-dose treatment, HFSR recurred in only one of these patients. SOR-induced nevi modification was not observed and no clear difference in ERK phosphorylation was reported between treated and non-treated patients. Dermatologic symptoms were easily managed with topical treatments.
Conclusions: Despite a high frequency of SOR-induced cutaneous toxicities, most were mild to moderate in severity and easily manageable. The majority of SOR-induced grade 2 HFSR can be treated symptomatically without dose decrease or interruption. For grade 3 HFSR, dose reduction (by 50%) or interruption for 5–7 days, plus symptomatic treatment, usually leads to rapid symptom relief. In this study, treatment discontinuation for HFSR was not necessary. Further experience at our center shows that even when discontinuation is necessary for severe HFSR, reinitiation at the same dose is often possible without reoccurrence of this grade of toxicity. Final clinical and pharmacodynamic results, including correlation of cutaneous events with patient response, will be reported.

106 POSTER

Methylnaltrexone inhibits S1P, VEGF and PDGF-induced angiogenesis: role of receptor transactivation

P. Singleton[1], J. Garcia[1], J. Moss[1], R. Israel[2]. [1]*University of Chicago, Medical Affairs, Chicago, USA;* [2]*Progenics Pharmaceuticals, Inc., Medical Affairs, Tarrytown, USA*

Background: The role of opioids in angiogenesis is largely unknown. The peripheral mu opioid antagonist, methylnaltrexone (MNTX), allows exploration of the role of opioids in angiogenesis. We have shown in human dermal microvascular endothelial cells (EC) that clinically relevant concentrations of morphine sulfate (MS) cause EC migration, inhibited by pretreatment with MNTX at therapeutically relevant levels (0.1 μM). To confirm the angiogenic effect of opioids and determine its mechanism, we examined MNTX in agonist-induced human pulmonary microvascular EC (HPMVEC) proliferation (P) and migration (M), two key elements in angiogenesis.

Methods: HPMVEC proliferation assay was performed as previously described. HPMVEC (5 * 103 cells/well) were incubated with serum-free media containing various agonists (100 nM MS, DAMGO or VEGF) for 24 h at 37ºC. The in vitro cell proliferation was analyzed by measuring increases in cell number using the CellTiter96TM MTS assay. 24 transwell units with 8 μM pore size were used for monitoring in vitro cell migration. HPMVEC (~1×10^4 cells/well) were plated with various treatments to the upper chamber and various agonists were added to the lower chamber. Cells were allowed to migrate for 18 hrs. Cells from the upper and lower chamber were counted by the same assay.
Results: S1P, VEGF, PDGF, MS and DAMGO induced P and M of EC which was inhibited by pretreatment with MNTX (0.1 μM, 1 hr). Silencing mu opioid receptor expression (siRNA) blocks MS and DAMGO-induced EC P and M while also inhibiting S1P, VEGF and PDGF-induced EC P and M. Immunoprecipitation followed by immunoblot indicate that S1P, VEGF and PDGF treatment of EC induced serine/threonine phosphorylation of the mu opioid receptor (indicating receptor transactivation) and activation of the G-protein, RhoA. MS and DAMGO treatment of EC induced tyrosine phosphorylation of the VEGF receptor, PDGF receptor and S1P3 receptor along with RhoA activation. MNTX pretreatment of EC attenuated MS, DAMGO, S1P, VEGF and PDGF-induced receptor phosphorylation events and RhoA activation. Finally, silencing RhoA expression blocked agonist-induced EC proliferation and migration.
Conclusion: These results indicate that MNTX inhibits agonist-induced EC P and M via inhibition of receptor phosphorylation/transactivation and subsequent inhibition of RhoA activation. MNTX inhibition of angiogenesis may be a useful therapeutic intervention for cancer treatment.
.

107 POSTER

Modulation of the radiation response of zebrafish embryos by targeting the VEGFR2 tyrosine kinase using ZD6474

G. Kari[1], A.Y. Zengin[2], B. Daroczi[1], A. Ryan[3], U. Rodeck[4], A.P. Dicker[1].
[1]Thomas Jefferson University, Radiation Oncology, Philadelphia, PA, USA; [2]Karadeniz Technical University Faculty of Medicine, Departments of Radiaiton Oncology, Trabzon, Turkey; [3]AstraZeneca, Manchaster, UK; [4]Thomas Jefferson University, Department of Dermatology, Philadelphia, PA, USA

Background: Preclinical studies have suggested that combining ionizing radiation with antiangiogenic agents enhances the therapeutic effect of ionizing radiation. Here, we addressed this issue *in vivo* using a novel vertebrate model, zebrafish embryos. Zebrafish are rapidly and prolifically bred and easily maintained, and embryos are optically transparent, facilitating direct observation of internal organs. Previously, we established zebrafish embryos as a model for the genotoxic stress response and pharmacologic modulation thereof (McAleer et al., *Int J Radiat Oncol Biol Phys* 61(1):10−13). The present study was designed to assess whether ZD6474 (AstraZeneca, Manchester UK), an inhibitor of VEGF receptor-2 (KDR) tyrosine kinase, modulated radiation sensitivity of zebrafish embryos.
Materials and Methods: Zebrafish were mated in embryo collection tanks. Viable embryos were washed and sorted at the one-cell developmental stage, and maintained under normoxic conditions at 28.5ºC for normal development. Morphology and survival was assessed visually using a light transmission microscope at 24-h intervals up to 144 hours post fertilization (hpf). The criterion for embryonic survival was the presence of cardiac contractility. Inhibition of angiogenesis was determined by monitoring the development of the main dorsal artery and intersegmental vessels.
Results: Treatment of live fish embryos with 10 μM ZD6474 at 24 hpf completely blocked formation of all blood vessels including the aorta as assessed at 48 hpf. At 3.3 μM ZD6474 approximately half (53%) of the embryos completely lacked vessel formation and none had developed intersegmental vessels, while at 1 μM only the development of the intersegmental vessels were perturbed (43%). When ZD6474 was administered within 30 min prior to ionizing radiation (0−20 Gy) at 24 hpf overall survival was markedly reduced. At 120 hours after irradiation only a fraction of the ZD6474-treated embryos (3.3 μM; 1.0 μM) were alive (10±5.8% and 34.8±14.7%, respectively) compared to 61.4±15.5% of control embryos receiving vehicle. Radiation-induced defects in midline development were significantly ($p < 0.05$) increased in ZD6474-treated irradiated embryos (93.3±5.8% and 82.8±13.4%, 3.3 μM; 1.0 μM respectively) vs. radiation alone (59.4±8.3%).
Conclusions: ZD6474 (10 μM) alone severely disturbed vascular development in zebrafish embryos. Concurrent administration of lower concentrations of ZD6474 and ionizing markedly reduced survival of zebrafish embryos, and sensitized them to radiation-induced morphological malformations. This model may help facilitate the evaluation of radiation modifiers.

108 POSTER

Phase I study of ABT869, a multiple receptor tyrosine kinase inhibitor, in patients with refractory solid malignancies.

B.C. Goh[1], C.H. Thng[2], N. Sukri[1], C.I. Wong[1], R. Soo[1], R. Carr[3], S.C. Lee[1], C.S. Chen[1], R. Humerickhouse[4]. *[1]National University Hospital, Department of Hematology-Oncology, Singapore, Singapore; [2]National Cancer Center, Department of Oncologic Imaging, Singapore; [3]Abbott Laboratories, Department of Clinical Pharmocokinetics, Abbott Park, USA; [4]Abbott Laboratories, Oncology Development, Abbott Park, USA*

ABT869 is an orally administered, potent and specific inhibitor of VEGF and PDGF family tyrosine kinases, including FLT-3, and c-kit receptors. A phase I study of ABT869 in patients with refractory solid malignancies was initiated to determine the maximum tolerable dose (MTD) of ABT869 given by continuous once-daily oral dosing in escalating doses; evaluate ABT869 pharmacokinetics; assess pharmacodynamic effects on plasma VEGF, flt-1 receptor, circulating endothelial cells and other potential biomarkers; and to evaluate tumor response, including an assessment of microcirculatory parameters (blood flow, F and capillary permeability, PS) with DCE-MRI. Dose escalation was planned in cohorts of 3 patients each. ABT869 was administered before bedtime except on days 1 and 15. Treatment periods (TP) were defined as 21 days and tumor assessments were performed using CT scans after every 6 weeks. DCE-MRI was done at baseline, day 3 and 14 of the first cycle. Cohort expansion to 6 patients was planned if dose limiting toxicity (DLT) occurred in the first cycle of treatment, and MTD was defined as the dose at which $\geqslant$2/6 patients experienced DLT. 4 male, 5 female patients (median age 55; range 29−73) have received a total of 34 TP; 6 at 10 mg per day and 3 at 0.25 mg/kg/day. Weight adjusted dosing was implemented to minimize interpatient variability. Cycle 1 toxicities included fatigue (grade 3 DLT in 1 patient at 10 mg), asthenia, myalgia (grade 2 in 4/9), skin rash (maculopapular, vasculitic in 1 patient), hand foot syndrome, hypertension, proteinuria and mouth irritation. Hypertension and proteinuria were reversible on dose interruption. Pharmacokinetics of ABT869 demonstrated plasma clearance of 2.8±1.3 L/h, with a corresponding mean half-life of 16±5 h. Drug accumulation was not significant with continuous dosing (day 15/day 1 accumulation ratio 1.16). The target AUC (4.9 μg h/mL) for activity based on preclinical models has been reached with daily dosing of 10 mg (mean 4.1±2.2 μg h/mL). A carboxylate-derivative was identified as a major metabolite, suggesting cytochrome p-450 enzymes play a role in ABT-869 metabolism. 5/6 patients at 10 mg achieved stable disease, with CT scan evidence of tumor necrosis and DCE-MRI evidence of reduced Ktrans, ve, F and PS. In conclusion, continuous dosing of ABT869 is tolerable and achieves target exposure at doses studied and demonstrates early DCE MRI evidence of reducing tumor flow and capillary permeability.

109 POSTER

Metronomic oral vinorelbine: dose escalation study, pharmacokinetics and assessment of predictive biomarkers

E. Briasoulis[1], K. Neanidis[2], P. Pappas[1], C. Tolis[2], C. Puozzo[3], M. Marselos[1], O. Siarabi[4], E. Tzamakou[2], G. Fountzilas[5], N. Pavlidis[1].
[1]University of Ioannina, Medical School, Ioannina, Greece; [2]University Hospital of Ioannina, Oncology, Ioannina, Greece; [3]Institut de Recherche Pierre Fabre, Pharmacokinetic Department, Castres, France; [4]Hellenic Cooperative Oncology Group, Data Management Unit, Ioannina, Greece; [5]University of Thessaloniki, Medical School, Thessaloniki, Greece

Background: Metronomic chemotherapy, the rhythmical dense administration of low doses of cytotoxics is believed to exert antitumor activity though damaging the activated endothelial cells in tumor vasculature. We investigated vinorelbine, an orally bioavailabe antimitotic drug, at a metronomic schedule of administration.
Material and Methods: patients with resistant metastatic tumors were treated with, escalated doses of oral vinorelbine, three times a week (TIW) without break until disease progression or unacceptable toxicity (UT) defined as any grade 2 toxicity according to CTC version 3. Patients were initially followed biweekly and later every month for assessing disease status, toxicity and blood sampling for pharmacokinetics and quantification of circulating angiogenesis related factors [VEGF, VEGFr2, TSP1, IL-8, FGFb and p53]. The study should close if UT occurred in two patients treated at minimum 3 months. The dose below this would be the highest metronomic dose (HMD).
Results: Eighty patients [39 women, median age 58, median PS 1] enrolled between June 2004 and December 2005 and treated at 7 dose levels: 20 mg (16 pts), 30 mg (17(, 40 mg (26), 50 mg (13), 60 mg (6) and 70 mg (2 pts). Median duration of treatment was 19 weeks (range 4 to 85+). Unacceptable toxicity occurred in 2 patients at the 60 mg dose level (leucopenia of grade 4 on 14th week of treatment and epistaxis on 9th week