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ORIGINAL REPORT

Phase II Study of Cediranib, an Oral Pan–Vascular Endothelial Growth Factor Receptor Tyrosine Kinase Inhibitor, in Patients With Recurrent Glioblastoma

Tracy T. Batchelor, Dan G. Duda, Emmanuelle di Tomaso, Marek Ancukiewicz, Scott R. Plotkin, Elizabeth Gerstner, April F. Eichler, Jan Drappatz, Fred H. Hochberg, Thomas Benner, David N. Louis, Kenneth S. Cohen, Houng Chea, Alexis Exarhopoulos, Jay S. Loeffler, Marsha A. Moses, Percy Ivy, A. Gregory Sorensen, Patrick Y. Wen, and Rakesh K. Jain

From the Stephen E. and Catherine Pappas Center for Neuro-Oncology, Massachusetts General Hospital Cancer Center; Departments of Neurology, Radiation Oncology, and Pathology; Center for Regenerative Medicine, Massachusetts General Hospital; Department of Neurology, Brigham and Women's Hospital; Center for Neuro-Oncology, Dana-Farber Cancer Institute; Vascular Biology Program, Children's Hospital Boston, Harvard Medical School, Boston; A.A. Martinos Center for Biomedical Imaging, Division of Health Sciences and Technology, Massachusetts Institute of Technology and Massachusetts General Hospital, Charlestown, MA; and Cancer Therapy Evaluation Program, National Cancer Institute, Bethesda, MD.

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A.G.S., P.Y.W., and R.J.K. contributed equally to this article.

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Corresponding author: Tracy T. Batchelor, MD, Stephen E. and Catherine Pappas Center for Neuro-Oncology, Yawkey 9E, Massachusetts General Hospital Cancer Center, 55 Fruit St, Boston, MA 02114; e-mail: tbatchelor@partners.org.

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ABSTRACT

Purpose
Glioblastoma is an incurable solid tumor characterized by increased expression of vascular endothelial growth factor (VEGF). We performed a phase II study of cediranib in patients with recurrent glioblastoma.

Methods
Cediranib, an oral pan-VEGF receptor tyrosine kinase inhibitor, was administered (45 mg/d) until progression or unacceptable toxicity to patients with recurrent glioblastoma. The primary end point was the proportion of patients alive and progression free at 6 months (APF6). We performed magnetic resonance imaging (MRI) and plasma and urinary biomarker evaluations at multiple time points.

Results
Thirty-one patients with recurrent glioblastoma were accrued. APF6 after cediranib was 25.8%. Radiographic partial responses were observed by MRI in 17 (56.7%) of 30 evaluable patients using three-dimensional measurements and in eight (27%) of 30 evaluable patients using two-dimensional measurements. For the 15 patients who entered the study taking corticosteroids, the dose was reduced (n = 10) or discontinued (n = 5). Toxicities were manageable. Grade 3/4 toxicities included hypertension (four of 31; 12.9%); diarrhea (two of 31; 6.4%); and fatigue (six of 31; 19.4%). Fifteen (48.4%) of 31 patients required at least one dose reduction and 15 patients required temporary drug interruptions due to toxicity. Drug interruptions were not associated with outcome. Changes in plasma placental growth factor, basic fibroblast growth factor, matrix metalloproteinase (MMP) -2, soluble VEGF receptor 1, stromal cell–derived factor-1α, and soluble Tek/Tie2 receptor and in urinary MMP-9/neutrophil gelatinase-associated lipocalin activity after cediranib were associated with radiographic response or survival.

Conclusion
Cediranib monotherapy for recurrent glioblastoma is associated with encouraging proportions of radiographic response, 6-month progression-free survival, and a steroid-sparing effect with manageable toxicity. We identified early changes in circulating molecules as potential biomarkers of response to cediranib. The efficacy of cediranib and the predictive value of these candidate biomarkers will be explored in prospective trials.

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INTRODUCTION

Despite treatment with surgery, radiation, and chemotherapy almost all patients with glioblastoma experience recurrence and the median survival for most patients is fewer than 15 months. Therapy with conventional and experimental agents for recurrent glioblastoma is unsatisfactory and the proportion of these patients who are alive and progression free at 6 months (APF6) is 9% to 15%.

Increased vascular permeability leading to cerebral edema and microvascular proliferation are hallmarks of glioblastoma.[1-4] This is due to high expression of proangiogenic cytokines, particularly of vascular endothelial growth factor (VEGF) and signaling via its endothelial tyrosine kinase receptor VEGF receptor 2 (VEGFR2).[5-7] Levels of VEGF and its receptor correlate with the histologic grade of gliomas.[8,9] We have previously shown that inhibiting the VEGF pathway normalizes the vasculature of

gliomas in preclinical models and in patients and that this vascular normalization extends survival in preclinical murine orthotopic models of glioblastoma.[10-13] Thus, recurrent glioblastoma has emerged as an attractive setting in which to conduct clinical trials of novel anti-VEGF agents, such as monoclonal antibodies (bevacizumab; Avastin, Genentech, South San Francisco, CA) or tyrosine kinase inhibitors (TKI; eg, cediranib, Recentin, AZD2171, AstraZeneca Pharmaceuticals, Cheshire, United Kingdom).[14]

Cediranib is an orally available pan-VEGFR tyrosine kinase inhibitor with a half-life of 22 hours compatible with once daily dosing.[15] Cediranib has a subnanomolar 50% inhibitory concentration for VEGF receptors with additional activity against platelet-derived growth factor ß and c-Kit. In a preliminary study in a subset of patients with recurrent glioblastoma, we observed that cediranib treatment normalizes tumor vasculature and alleviates edema.[10] Herein, we report the final clinical efficacy, toxicity, and biomarker data on the entire cohort of patients treated on the first phase II study of cediranib in recurrent glioblastoma.

METHODS

Study Design

This phase II study of cediranib was approved by the local institutional review board (IRB) and was sponsored by the National Cancer Institute (NCI, NCT00305656). All patients signed an IRB-approved informed consent document before enrollment. The primary end point of this study was APF6, and secondary end points included radiographic response proportion, median overall survival (OS), and toxicity. Inclusion criteria for patients included pathologic diagnosis of glioblastoma; age $\geq$ 18 years; Karnofsky performance score $\geq$ 60; Mini-Mental Status Examination score $\geq$ 15; prior therapy with radiation; treatment with $\leq$ 2 chemotherapy regimens; recurrent glioblastoma by magnetic resonance imaging (MRI) or by tissue diagnosis; stable dose of corticosteroids for $\geq$ 5 days before the first baseline MRI scan; elapse of $\geq$ 3 months since completion of radiation; elapse of $\geq$ 3 weeks since completion of a non-nitrosourea chemotherapy; elapse of $\geq$ 6 weeks since completion of a nitrosourea-based chemotherapy; adequate bone marrow function (absolute neutrophil count $\geq$ 1,500/mcl; hemoglobin $\geq$ 8g/dL; platelet count $\geq$ 100,000/mcl); creatinine within institutional normal limit or creatinine clearance $\geq$ 60 mL/min/1.73 m^2 for patients with creatinine more than institutional normal limits. Exclusion criteria included major surgery (including craniotomy) $\leq$ 4 weeks before the start of cediranib; concurrent use of anticoagulants; concurrent use of enzyme-inducing antiepileptic drugs; mean corrected QT interval more than 470 milliseconds or patients with a history of familial prolonged QT syndrome; $\geq$ 1 proteinuria on two consecutive urine dipstick assessments; pregnancy; history of uncontrolled hypertension or other serious medical illnesses including, but not limited to, unstable angina, arrhythmia, symptomatic congestive heart failure, active infection; infection with the human immunodeficiency virus; imaging (computed tomography or MRI) evidence of intratumoral or intracerebral hemorrhage deemed significant by the treating physician.

All patients were initially treated with cediranib 45 mg once each day. A cycle was defined as 28 days. A step-wise dose reduction scheme (Starting dose: 45 mg$\rightarrow$dose level $-$1: 30 mg$\rightarrow$dose level $-$2: 20 mg$\rightarrow$dose level $-$3: 10 mg) was utilized in patients who experienced dose-limiting toxicities. Patients were also allowed to temporarily interrupt cediranib for toxicity and resume the drug up to 14 days later. Algorithms for management of hypertension and diarrhea were followed when these toxicities were observed.

Treatment Response Evaluation

All patients were monitored by serial physical examinations, laboratory tests, and MRI scans. The MRI sequences included T1 pre-/postcontrast, T2, fluid-attenuated inversion recovery (FLAIR), diffusion weighted imaging, perfusion weighted imaging (dynamic susceptibility contrast) and dynamic contrast enhanced imaging. A schedule of the laboratory tests and MRI scans is enumerated in Appendix Table A1 (online only). Two baseline MRI scans were obtained 1 to 7 days before the first dose of cediranib followed by another MRI scan within 24 hours after the first dose of the medication then every month thereafter. The second baseline MRI scan (closer to the initiation of treatment) was used as the baseline for comparison of all subsequent studies. The postcontrast, T1-weighted MRI scans were assessed for response using a volumetric program by a central neuroradiologist who was blinded to patient identity and date of the scan. Scans were presented for review in a randomized sequence. The MRI scans were also assessed with two-dimensional measurements based on published criteria.[16] Disease progression was defined according to Macdonald criteria. An independent radiologist from the Cancer Therapy Evaluation Program at the National Cancer Institute also confirmed radiographic responses in patients enrolled at the halfway point of the study. All toxicities were reported according to the National Cancer Institute Common Toxicity Criteria, version 3.

Circulating Biomarker Evaluations

Peripheral blood was obtained from all patients before therapy then 8 hours, 1 day, 9 days, 28 days, 56 days, 84 days, and 112 days thereafter to measure circulating proangiogenic and proinflammatory molecules and cells. Circulating progenitor cells were enumerated by flow cytometry using CD31, CD34, CD45, and CD133 as markers.[17] Plasma analysis was carried out for circulating VEGF, placental growth factor (PlGF), sVEGFR1, basic fibroblast growth factor (bFGF), interleukin (IL) -1β, IL-6, IL-8, transforming growth factor α, matrix metalloproteinase (MMP) -2, and MMP-10 using multiplex enzyme-linked immunosorbent assay plates from Meso-Scale Discovery (Gaithersburg, MD) as well as soluble VEGFR2, stromal cell–derived factor-1α (SDF1α), angiopoietin 1 (Ang1), angiopoietin 2 (Ang2), and soluble Tek/Tie2 receptor (sTie2) from R&D System (Minneapolis, MN). Every sample was run in duplicate. Urine samples were obtained at similar time points as used for blood collection from the last 15 consecutive patients. Urinary MMP-2 (65kDa), MMP-9 (95kDa), and MMP-9/neutrophil gelatinase-associated lipocalin (NGAL) complex (125kDA) and activity were evaluated using gel zymography and were semi-quantitatively assessed by scoring from 1 (absent) to 9 (very strong).[18-20]

Data and Statistical Analysis

Published historical outcomes in recurrent glioblastoma report an APF6 of 9% to 15%, median progression-free survival (PFS) of 54 to 63 days and median OS of 150 to 175 days.[21,22] This phase II study was designed to detect an increase in APF6 from 10% to 25%.

Changes from baseline MRI parameters or circulating biomarkers were analyzed using the paired exact Wilcoxon test. *P* values were adjusted for multiple comparisons using Hommel's method.

Univariate analyses of PFS and OS with sex, age, Karnofsky performance status, baseline circulating, or urinary biomarkers and their early changes at 8 hours and 1 day were performed using a Wald test in the Cox proportional hazards model. Biomarker levels measured on quantitative scales were log-transformed and changes were calculated as ratios of on-study to baseline values. Analysis of the effect of drug interruptions on PFS and OS was performed using a Wald test in the time-dependent proportional hazards model, adjusting for the Vascular Normalization Index[12] and using a sandwich estimator of variance[23] to account for correlated data within patients.

Finally, we performed correlation analyses between all MRI-measured T1-contrast–enhanced tumor volumes and levels of plasma proteins and cell biomarkers at corresponding time points. This analysis of potential biomarkers of response and recurrence was based on a mixed-effects model, using the log-transformed biomarker level and a B-spline function of time in the fixed-effects model part and patient-specific linear function of time (including intercept) in the random-effect part.

RESULTS

Patient Characteristics

The study enrolled 31 patients with recurrent glioblastoma who had experienced prior treatment failure (Table 1). One patient who

Table 1. Patient Characteristics

Parameter	Value
Median age, years	53
Range	20-77
Median Karnofsky performance score	90
Range	70-100
Sex (male:female)	18:13
Initial surgery	
Biopsy	5
Resection	26
Prior temozolomide	29
No. of prior chemotherapies	
1	24
> 1	7
Median dexamethasone dose at study entry, mg (n = 15)	8
Range	1-16
History of hypertension	6

received only 18 doses of cediranib was included in the assessment of the OS, PFS, APF6, and toxicity, but excluded from other analyses. Eighteen of 31 patients had been treated by prior partial or total resection, 31 of 31 patients received prior radiation, and 29 of 31 patients had received prior temozolomide. Fifteen patients entered the study on dexamethasone with a median dose of 8 mg daily.

Radiographic Tumor Response, Radiographic Disease Progression, and OS

All patients eventually experienced tumor progression and died except for one patient who remains alive having experienced disease progression after 26 months of cediranib therapy. The proportion of patients who achieved a partial radiographic response (> 50% reduction in contrast-enhancing volume) after treatment with cediranib was 56.7% using volumetric criteria and 27% using Macdonald criteria (Table 2). The APF6 was 25.8%, the median PFS was 117 days, and the median OS was 227 days (Table 2). Three patients were removed from the study by their treating physicians due to clinical progression without radiographic progressive disease. An independent review of MRI scans at the halfway point of the study confirmed all radiographic responses. Five patients demonstrated more than 25% increase in FLAIR dimensions 1 to 2 months before the observation of progressive disease on the postcontrast T1-weighted MRI sequences. None of these five patients was removed from the study due to clinical progression.

Partial (over 50% volume reduction; n = 17) or minor responses (25% to 50% volume reduction; n = 6) correlated significantly with PFS ($P < .05$) but not with OS. Age was associated with a higher hazard of death ($P = .027$, Wald test), and Karnofsky performance status correlated with best radiographic responses after treatment ($\rho = -0.51$; $P = .004$).

Fifteen patients entered the study on a dose of corticosteroids. After cediranib treatment, the dose was reduced in 10 of 15 of these patients and corticosteroids were discontinued in five of 15 patients. Conversely, after discontinuation of cediranib 18 of 29 patients required either initiation of dexamethasone or a higher dose of dexamethasone. All cranial MRI sequences related to vasogenic cerebral edema (FLAIR, apparent diffusion coefficient, extracellular-extravascular volume fraction) demonstrated significant reductions after administration of cediranib and these changes persisted for at least 1 cycle (28 days, Appendix Table A1).

Table 2. Efficacy of Cediranib in Recurrent Glioblastoma

Radiographic Response	Cediranib (AZD2171)		Wong et al[20]	
	No.	%	No.	%
Volumetric criteria				
Partial	17/30	56.7	NA	
Minor	6/30	20.0		
Macdonald criteria				
Partial	8/30	26.6	NA	
APF6 (%)	31	25.8		15
95% CI	14.7 to 46.9			
PFS, days	117 (N = 31)		63	
95% CI	82 to 145			
OS, days	227 (N = 31)		175	
95% CI	177 to 293			

Abbreviations: NA, not applicable; APF6, alive and progression free at 6 months; PFS, progression-free survival; OS, overall survival.

Safety and Tolerability

Two patients elected to stop the treatment due to fatigue. There were no other study terminations due to toxicity and there were no treatment-related deaths. There were no intratumoral or intracerebral hemorrhages observed during this study. The most common toxicities observed were hypertension, fatigue, and diarrhea. Grade 3/4 toxicities considered as possibly, probably, or definitely related to cediranib were observed in 21 (68%) of 31 of patients and are summarized in Table 3. Fifteen of 31 patients required at least one dose reduction

Table 3. National Cancer Institute Common Toxicity Criteria Grade 3 or 4 Toxicities Possibly, Probably, or Definitely Related to Cediranib

Toxicity	Frequency (N = 31)	
	No.	%
Fatigue	6	19
ALT	5	16
Hypertension	4	13
Abdominal pain	4	13
AST	3	10
Diarrhea	2	6
Bilirubin	2	6
Hypophosphatemia	2	6
Metabolic/laboratory, other	2	6
Lower extremity weakness	2	6
Headache	2	6
Leukocytes	1	3
Neutrophils	1	3
Platelets	1	3
Hand-foot reaction	1	3
Ulceration	1	3
Obstruction, gallbladder	1	3
Hypokalemia	1	3
Proteinuria	1	3
Memory impairment	1	3
Depressed level of consciousness	1	3
Thrombosis	1	3

Table 4. Plasma Cytokines (pg/mL) That Significantly Change After Cediranib Treatment in Patients With Recurrent Glioblastoma

	Pretreatment					8 Hours					Day 1					Day 9				
Biomarker	Median	Interquartile Range	No.	*P*	P_{adj}*	Median	Interquartile Range	No.	*P*	P_{adj}*	Median	Interquartile Range	No.	*P*	P_{adj}*	Median	Interquartile Range	No.	*P*	P_{adj}*
Plasma VEGF	139	115-183	31	NA	NA	**176**	**137-231**	**31**	**< .001**	**< .001**	**212**	**167-323**	**31**	**< .001**	**< .001**	**281**	**219-372**	**30**	**< .001**	**< .001**
Plasma PlGF	21	17,25	31	NA	NA	**35**	**30-45**	**31**	**< .001**	**< .001**	**58**	**40,77**	**31**	**< .001**	**< .001**	**79**	**47,128**	**30**	**< .001**	**< .001**
Plasma SDF1α	1,602	1,301-1,977	30	NA	NA	**1,621**	**1,344-2,169**	**30**	**< .001**	**< .001**	**1,729**	**1,491-2,338**	**31**	**< .001**	**< .001**	**1,886**	**1,326-2,240**	**30**	**< .001**	**.068**
Plasma sVEGFR2	7,917	6,556-10,334	31	NA	NA	8,051	6,226-10,026	31	0.72	0.72	8,188	6,147-10,062	31	0.42	0.72	*6,854*	*5,068-8,773*	*30*	*< .001*	*< .001*
Plasma sTie2	9,970	7,770-12,085	31	NA	NA	9,650	7,785-11,057	31	.49	.69	10,050	7,960-12,330	31	.69	.69	*8,610*	*6,670-9,862*	*30*	*< .001*	*< .001*
Plasma MMP-2	1,378	855-1,937	25	NA	NA	1,317	1,106-1,569	25	.77	.98	*1,157*	*809-1,507*	*25*	*< .001*	*.020*	1,135	737-1,594	25	.034	.30
Plasma MMP-10	1.01	0.74-1.55	31	NA	NA	*0.94*	*0.64-1.31*	*31*	*< .001*	*< .001*	1.02	0.73-1.34	31	.15	.15	1.25	0.89,1.53	31	.14	.15
Plasma Ang2	1,532	1,229-1,858	31	NA	NA	1,532	1,280-1,940	31	.84	.84	1,485	1,161-1,923	31	.23	.54	*1,248*	*1,038-1,640*	*30*	*< .001*	*.018*

(continued on following page)

NOTE. *P* indicates values that are from the paired exact Wilcoxon tests, unadjusted. Bold font indicates increase; italic font indicates decrease.
Abbreviations: VEGF, vascular endothelial growth factor; PlGF, placental growth factor; SDF1α, stromal cell–derived factor-1α; sVEGFR2, soluble VEGF receptor 2; sTie2, soluble Tek/Tie2 receptor; MMP, matrix metalloproteinase; Ang2, angiopoietin 2.
*P_{adj} values are from the paired exact Wilcoxon tests, adjusted to control the false discovery rate over time, with weights proportional to the square root of the number of the measurements.

while on the study treatment due to toxicity and 15 of 31 patients required a temporary drug interruption due to toxicity. The most common reasons for interruption were diarrhea (n = 3), hypertension (n = 2), proteinuria (n = 2), low thyroid stimulating hormone (n = 2), and hand-foot syndrome (n = 2). Twenty-seven of 31 patients treated with cediranib developed ≥ grade 1 hypertension after initiation of cediranib and 25 of 31 patients required medical treatment for hypertension. Drug interruptions had no significant association with mortality or disease progression ($P > .8$).

The hazard of disease progression correlated inversely with diarrhea grade ($P = .004$, Wald test), but not with the hypertension grade ($P = .18$, Wald test). There were no significant correlations between these toxicities and OS.

Circulating Biomarker Analysis

In line with previous findings,[10] biomarker kinetics after cediranib treatment in patients with recurrent glioblastoma were associated with immediate (by 8 hours) and persistent elevations in plasma of PlGF, SDF1α, and VEGF and more delayed decreases in soluble VEGFR2 (sVEGFR2; ie, by day 9; Table 4). In addition, we observed that cediranib treatment induced an immediate and persistent increase in MMP-10, a more delayed but persistent decrease in sTie2, and transient decreases in MMP-2 and Ang2 in plasma ($P < .01$; Table 4). The levels of VEGF, PlGF, and MMP-10 significantly decreased, and those of sVEGFR2 and sTie2 significantly increased after cediranib interruptions (ie, when measured within 2 weeks of the drug interruption). In this cohort, we detected no significant trends for the kinetics after treatment of circulating progenitor cells, or in plasma levels of bFGF, sVEGFR1, Ang1, IL-1β, IL-6, IL-8 or transforming growth factor α, and urinary MMP-2, MMP-9 or MMP-9/NGAL activity (Appendix Tables A2-A4 online only).

The association between cediranib treatment outcome measures (OS, PFS) and biomarkers was explored for baseline levels as well as for early changes in these biomarkers. None of the biomarkers showed

Table 5. Association Between Blood Angiogenic Biomarkers at Baseline, Their Changes at 8 Hours and at Day 1 After Cediranib Treatment With Radiographic Progression of Disease, and Mortality in Patients With Recurrent Glioblastoma

	Pretreatment Measurement						Change at 8 Hours		
	Progression			Mortality			Progression		
Biomarker	Estimate	95% CI	No. of Observations	Estimate	95% CI	No. of Observations	Estimate	95% CI	No. of Observations
Plasma PlGF	−11	66 to 130	30	21	−48 to 182	30	−22	−67 to 85	30
P		.80			.66			.56	
Plasma bFGF	−16	−33 to 6	30	−13	−29 to 7	30	−16	−41 to 20	30
P		.18			.21			.34	
Plasma MMP-2	74	−1 to 206	25	−20	−52 to 36	25	**351**	**20 to 1,951**	**25**
P		.057			.41			**.020**	
Urinary MMP-9/NGAL activity	0	−15 to 18	12	2	−13 to 20	12	14	−14 to 50	11
P		.97			.81			.35	

(continued on following page)

NOTE. Data are shown as estimates of the association with PFS or OS (with 95% CIs), the No. of observations, and *P* values for the likelihood ratio test. Except for urinary protein activity, the estimates are percent increases of the hazard ratio (decreases, for negative estimates) corresponding to a doubling of the biomarker value. For urinary proteins the estimates are percent increases of the hazard ratio (decreases, for negative estimates) corresponding to an increase of the marker level by one category. Statistically significant correlations are marked by asterisks. The changes were modeled using measurements at a given time point and adjusting for baseline levels. Bold font indicates decrease.
Abbreviations: PlGF, placental growth factor; bFGF, basic fibroblast growth factor; MMP, matrix metalloproteinase; NGAL, neutrophil gelatinase-associated lipocalin.

Table 4. Plasma Cytokines (pg/mL) That Significantly Change After Cediranib Treatment in Patients With Recurrent Glioblastoma (continued)

Biomarker	Day 28 Median	Day 28 Interquartile Range	Day 28 No.	Day 28 P	Day 28 P_{adj}*	Day 56 Median	Day 56 Interquartile Range	Day 56 No.	Day 56 P	Day 56 P_{adj}*	Day 84 Median	Day 84 Interquartile Range	Day 84 No.	Day 84 P	Day 84 P_{adj}*	Day 112 Median	Day 112 Interquartile Range	Day 112 No.	Day 112 P	Day 112 P_{adj}*
Plasma VEGF	**236**	**197-461**	**30**	**< .001**	**< .001**	**282**	**223-594**	**22**	**< .001**	**< .001**	**455**	**344-535**	**17**	**< .001**	**< .001**	**536**	**302-712**	**16**	**< .001**	**< .001**
Plasma PlGF	**69**	**49-121**	**30**	**< .001**	**< .001**	**94**	**59-169**	**22**	**< .001**	**< .001**	**135**	**100-185**	**17**	**< .001**	**< .001**	**161**	**70-244**	**16**	**< .001**	**< .001**
Plasma SDF1α	1,694	1,479-2,150	30	.087	.26	1,682	1,376-2,446	22	.079	.26	1,731	1,359-2,436	17	.020	.14	1,550	1,195-2,384	16	.13	.33
Plasma sVEGFR2	*5,486*	*3,986-6,451*	*30*	*< .001*	*< .001*	*4,950*	*4,044-6,717*	*22*	*< .001*	*< .001*	*4,276*	*3,404-5,551*	*17*	*< .001*	*< .001*	*4,338*	*3607,5823*	*16*	*< .001*	*< .001*
Plasma sTie2	*8,120*	*7,002-10,540*	*30*	*< .001*	*< .001*	*7,810*	*7,271-9,964*	*22*	*< .001*	*.022*	*7,690*	*7,100-10,100*	*8*	*< .001*	*< .001*	*7,565*	*7,085-9,434*	*14*	*< .001*	*.012*
Plasma MMP-2	1,183	789-1,590	25	.32	.98	1,172	920-1,344	19	.41	.98	1,108	990-1,627	15	.98	.98	1,188	937-1,977	10	.84	.98
Plasma MMP-10	**1.80**	**1.06-2.52**	**30**	**< .001**	**< .001**	**1.98**	**1.18-4.23**	**22**	**< .001**	**< .001**	**2.45**	**1.73,3.38**	**17**	**< .001**	**< .001**	**3.55**	**1.96-6.95**	**16**	**< .001**	**< .001**
Plasma Ang2	1,226	1,008-1,660	30	.016	.14	1,341	952-2,106	22	.17	.53	1,409	1,037-1,757	17	.043	.34	1,346	1,090-1,714	14	.17	.52

correlations with PFS or OS when evaluated at baseline. However, several dynamic biomarkers showed significant correlations with outcome. An increase in plasma MMP-2 at 8 hours after first administration of cediranib correlated with reduced PFS and OS ($P < .05$, Table 5). When measured at 1 day after treatment, an increase in urinary MMP-9/NGAL activity was associated with poor PFS ($P < .01$), and the extent of increase in PlGF and bFGF was significantly associated with longer OS ($P < .05$, Table 5). No other early biomarker changes correlated with OS or PFS (Appendix Table A5, online only).

We also evaluated the correlation between biomarker changes at any time point during treatment and radiographic response in individual patients. A radiographic PR (ie, decreases of $> 50\%$ in enhancing tumor volume) was significantly associated with higher levels of plasma PlGF and IL-8 and lower levels of bFGF and sTie2 measured at the same time-point ($P < .05$). In addition, radiographic tumor progression (ie, increases of $> 25\%$ in enhancing tumor volume) was significantly correlated with increased levels of sVEGFR1, sTie2, and SDF1α ($P < .05$).

DISCUSSION

Bevacizumab—a humanized monoclonal antibody that specifically targets VEGF-A ligand—was approved by the US Food and Drug Administration as monotherapy for recurrent glioblastoma based on two phase II studies. In a noncomparative, randomized phase II trial of bevacizumab alone versus bevacizumab and irinotecan in patients with recurrent glioblastoma there were radiographic overall response rates of 28.2% and 37.8% and APF6 proportions of 42.6% and 50.3%, respectively.[24] In another single-arm phase II study of bevacizumab alone followed by bevacizumab with irinotecan at progression in 48 patients with recurrent glioblastoma the APF6 was 29%.[25]

Based on the initial promising results with bevacizumab, several studies of oral agents that inhibit VEGF signaling have been conducted in the recurrent glioblastoma patient population with mixed results. A study of vatalanib (Novartis, Basel, Switzerland)—another oral pan-VEGFR TKI with additional activity against platelet-derived growth factor ß—with chemotherapy showed that fewer than 10% of the patients with recurrent glioblastoma achieved radiographic responses with a once-daily dosing schedule.[26] Herein we report the first phase II trial of oral cediranib for recurrent glioblastoma. Potential advantages of cediranib relative to bevacizumab include oral bioavailability; a shorter half-life (22 hours *v* 21 days), which should allow more rapid clearance of drug in the event of serious toxicity such as hemorrhage; multiple tyrosine kinase targets and the ability to target intracellular VEGF receptors. We observed that cediranib treatment results in a radiographic response proportion, APF6 proportion, median PFS and median OS that compare favorably with data from historical controls.[21] These data are also comparable to data obtained in phase II studies of bevacizumab in this patient population.[27] The frequency of drug discontinuation due to toxicity was low and

Table 5. Association Between Blood Angiogenic Biomarkers at Baseline, Their Changes at 8 Hours and at Day 1 After Cediranib Treatment With Radiographic Progression of Disease, and Mortality in Patients With Recurrent Glioblastoma (continued)

Biomarker	Mortality Estimate	Mortality 95% CI	Mortality No. of Observations	Change at Day 1: Progression Estimate	Progression 95% CI	Progression No. of Observations	Change at Day 1: Mortality Estimate	Mortality 95% CI	Mortality No. of Observations
Plasma PlGF	−47	−79 to 32	30	−37	−67 to 20	30	**−52**	**−76 to −5**	**30**
P		.17			.14			**.025**	
Plasma bFGF	−15	−39 to 17	30	−14	−43 to 29	30	**−35**	**−56 to −2**	**30**
P		.32			.47			**.040**	
Plasma MMP-2	**310**	**12 to 1,403**	**25**	186	−22 to 946	25	127	−38 to 724	25
P		**.031**			.094			.19	
Urinary MMP-9/NGAL activity	51	−7 to 144	11	**664**	**57 to 3,620**	**11**	40	−25 to 163	11
P		.059			**.0022**			.32	

comparable to other anti-VEGF therapies. The safety profile of cediranib in patients with glioblastoma was acceptable, and there were no CNS hemorrhages or increased risk of thromboembolic complications.

Radiographic assessments of tumor response and progression to anti-VEGF therapies are challenging as these agents reduce permeability and, consequently, contrast leakage.[28] Alternative radiographic methods are under investigation in order to more accurately define tumor response and progression in the setting of these agents. In this phase II trial progression of FLAIR signal abnormality was noted in five (16.6%) of 30 subjects before the observation of progressive disease on postcontrast T1-weighted sequences. This routinely acquired MRI sequence, as well as others including diffusion sequences, may therefore offer additional insight into disease progression in this patient population.[29]

The mechanism(s) of action of cediranib in patients with recurrent glioblastoma remains unclear. Cediranib treatment can transiently normalize the tumor vasculature and alleviate tumor-induced cerebral edema.[10,30] Normalization of glioblastoma vessels may reduce tumor hypoxia and enhance sensitivity to concurrently administered cytotoxic therapies including ionizing radiation and chemotherapy. Thus, there is a strong rationale to test cediranib in combination with chemotherapy and radiation in patients with newly diagnosed glioblastoma. The antiedema effect and consequent reduction in corticosteroid use also has the potential to provide clinical benefit to patients with glioblastoma. In addition, as observed in preclinical models of glioblastoma treated with cediranib, edema alleviation may result in prolonged survival even without inhibition of tumor growth.[11] Another potential antitumor mechanism could be targeting of the stem cell-like cancer cells in glioblastoma.

A major issue remains the heterogeneity in recurrent glioblastoma responses to cediranib, as observed for other anti-VEGF agents in various tumors.[14] To date, there are no validated biomarkers of response to anti-VEGF therapy. Thus, identifying biomarkers that may predict benefit versus lack of benefit early during the treatment course is highly desirable.

We evaluated multiple plasma molecules and circulating cells that have been implicated in tumor angiogenesis.[1] Biomarker kinetics were consistent with data on anti-VEGFR TKIs in our prior reports and others.[10,31,32] In line with published literature, the baseline levels of any of these biomarkers did not appear to predict response.[32,33] However, several of the biomarkers evaluated in our study (VEGF, PlGF, MMP-10, sVEGFR2, sTie2) changed significantly and reversibly after VEGF blockade. These are potential pharmacodynamic biomarkers, as similar changes have been reported for cediranib, vatalanib, and sunitinib in glioblastoma and other cancers.[34-39] Moreover, we observed significant correlations between several dynamic biomarkers (ie, the early change in plasma MMP-2, PlGF, sTie2, bFGF, and urinary MMP-9/NGAL activity) and radiographic responses and survival in recurrent glioblastoma after cediranib treatment. Increases in SDF1α, sVEGF1, and sTie2 were observed in patients at the time of glioblastoma progression after cediranib treatment. These observations are consistent with biomarker data from studies of sunitinib in hepatocellular carcinoma and bevacizumab in rectal cancer,[39,40] and should be validated in preclinical and larger clinical studies.

Another important issue raised by some investigators is the potential of increasing the frequency of disease progression after interruption of anti-VEGF therapies, as seen in some mouse models.[41,42] However, in this phase II study, there was no association of tumor progression with drug interruption.

In conclusion, cediranib monotherapy is active against recurrent glioblastomas and is associated with manageable toxicity. Further studies are warranted to confirm these results and to optimize the use of cediranib alone or in combination with cytotoxic therapies in patients with recurrent or newly diagnosed glioblastoma. Along these lines a randomized, three-arm, placebo-controlled, phase III trial in recurrent glioblastoma to test the efficacy of cediranib in this patient population has been initiated as well as studies of cediranib in combination with chemotherapy and radiation for patients with newly diagnosed glioblastoma.

AUTHORS' DISCLOSURES OF POTENTIAL CONFLICTS OF INTEREST

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AUTHOR CONTRIBUTIONS

Conception and design: Tracy T. Batchelor, Dan G. Duda, Emmanuelle di Tomaso, Marek Ancukiewicz, Jay S. Loeffler, Percy Ivy, A. Gregory Sorensen, Patrick Y. Wen, Rakesh K. Jain

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Provision of study materials or patients: Tracy T. Batchelor, Dan G. Duda, Emmanuelle di Tomaso, Scott R. Plotkin, April F. Eichler, Jan Drappatz, Fred H. Hochberg, David N. Louis, Kenneth S. Cohen, Alexis Exarhopoulos, Jay S. Loeffler, Marsha A. Moses, A. Gregory Sorensen, Patrick Y. Wen, Rakesh K. Jain

Collection and assembly of data: Tracy T. Batchelor, Dan G. Duda, Emmanuelle di Tomaso, Marek Ancukiewicz, Scott R. Plotkin, Elizabeth

Gerstner, April F. Eichler, Jan Drappatz, Fred H. Hochberg, Thomas Benner, David N. Louis, Kenneth S. Cohen, Houng Chea, Alexis Exarhopoulos, Marsha A. Moses, Percy Ivy, A. Gregory Sorensen, Patrick Y. Wen, Rakesh K. Jain

Data analysis and interpretation: Tracy T. Batchelor, Dan G. Duda, Emmanuelle di Tomaso, Marek Ancukiewicz, Scott R. Plotkin, Elizabeth Gerstner, April F. Eichler, Jan Drappatz, Fred H. Hochberg, Thomas Benner, David N. Louis, Kenneth S. Cohen, Houng Chea, Alexis Exarhopoulos, Jay S. Loeffler, Marsha A. Moses, Percy Ivy, A. Gregory Sorensen, Patrick Y. Wen, Rakesh K. Jain

Manuscript writing: Tracy T. Batchelor, Dan G. Duda, Emmanuelle di Tomaso, Marek Ancukiewicz, Scott R. Plotkin, Elizabeth Gerstner, April F. Eichler, David N. Louis, Percy Ivy, A. Gregory Sorensen, Patrick Y. Wen, Rakesh K. Jain

Final approval of manuscript: Tracy T. Batchelor, Dan G. Duda, Emmanuelle di Tomaso, Marek Ancukiewicz, Scott R. Plotkin, Elizabeth Gerstner, April F. Eichler, Jan Drappatz, Fred H. Hochberg, Thomas Benner, David N. Louis, Kenneth S. Cohen, Houng Chea, Alexis Exarhopoulos, Jay S. Loeffler, Marsha A. Moses, Percy Ivy, A. Gregory Sorensen, Patrick Y. Wen, Rakesh K. Jain

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