Neuro-Oncology 14(12):1503–1510, 2012.
doi:10.1093/neuonc/nos256
Advance Access publication October 22, 2012

EORTC 26083 phase I/II trial of dasatinib in combination with CCNU in patients with recurrent glioblastoma

Enrico Franceschi, Roger Stupp, Martin J. van den Bent, Carla van Herpen, Florence Laigle Donadey, Thierry Gorlia, Monika Hegi, Benoit Lhermitte, Lewis C. Strauss, Anouk Allgeier, Denis Lacombe, and Alba A. Brandes

Department of Medical Oncology, Bellaria-Maggiore Hospital, Azienda USL of Bologna, Italy (E.F., A.A.B.); University of Lausanne Hospitals, Lausanne, Switzerland (R.S.); Neuro-Oncology Unit, Department of Neurology, Daniel den Hoed Cancer Center/Erasmus Medical Center, Rotterdam, The Netherlands (M.J.v.d.B.); Departments of Medical Oncology, Radboud University Nijmegen Medical Centre, Nijmegen, The Netherlands (C.v.H.); Service de Neurologie Mazarin, Hôpital de la Pitié-Salpêtrière, Paris, France (F.L.D.); European Organisation for Research and Treatment of Cancer (EORTC) Headquarters, Brussels, Belgium (T.G., A.A., D.L.); Laboratory of Brain Tumor Biology and Genetics, University Hospital Lausanne (CHUV) and University of Lausanne, Lausanne, Switzerland (M.H.); Department of Pathology, Lausanne University Hospital, Lausanne, Switzerland (B.L.); Bristol-Myers Squibb Inc., Wallingford, Connecticut (L.C.S.)

The treatment of patients with recurrent glioblastoma remains a major oncologic problem, with median survival after progression of 7–9 months. To determine the maximum tolerated dose and dose-limiting toxicity (DLT), the combination of dasatinib and cyclonexyl-chloroethyl-nitrosourea (CCNU) was investigated in this setting. The study was designed as multicenter, randomized phase II trial, preceded by a lead-in safety phase. The safety component reported here, which also investigated pharmacokinetics and preliminary clinical activity, required expansion and is therefore considered a phase I part to establish a recommended dosing regimen of the combination of CCNU (90–110 mg/m^2) and dasatinib (100–200 mg daily). Overall, 28 patients were screened, and 26 patients were enrolled. Five dose levels were explored. DLTs, mainly myelosuppression, occurred in 10 patients. Grade 3 or 4 neutropenia was recorded in 7 patients (26.9%) and thrombocytopenia in 11 patients (42.3%). No significant effect of CCNU coadministration on dasatinib pharmacokinetics was found. Median progression-free survival (PFS) was 1.35 months (95% confidence interval: 1.2–1.4) and 6-month PFS was 7.7%. In this phase I study of recurrent glioblastoma patients, the combination of CCNU and dasatinib showed significant hematological toxicities and led to suboptimal exposure to both agents.

Keywords: CCNU, dasatinib, recurrent glioblastoma.

Prognosis of patients with glioblastoma remains poor, with a median survival of approximately 15 months and a 2-year survival rate of 27%, despite surgical resection, temozolomide chemotherapy, and radiation therapy (RT).[1] The majority of patients have recurrence within 1 year and usually die of the disease within 6–8 months after recurrence. Once a glioblastoma recurs, treatment options are limited, and chemotherapy is commonly proposed. Novel targeted approaches have therefore been evaluated in this setting.[2–4] The central role of the Src-kinase family in the pathogenesis of glioblastoma has recently been thoroughly documented,[5,6] and it is known that Src is responsible for the invasiveness of glioblastoma cells.[7,8] The Src-kinase family consists of 9 related nonreceptor tyrosine kinases, several of which are implicated in cancer, regulating 3 main cellular functions: adhesion, invasion, and motility. Dasatinib (BMS [Bristol-Myers Squibb]-354825) is a potent orally available inhibitor of Src kinases and has been approved for the treatment of chronic myeloid leukemia. Dasatinib also inhibits

Received June 6, 2012; accepted August 27, 2012.

Corresponding author: Alba A. Brandes, MD, Department of Medical Oncology, Bellaria-Maggiore Hospital, Via Altura 3, 40139 Bologna Italy (alba.brandes@yahoo.it).

several other oncogenic kinases linked to diverse malignancies, including c-Kit and c-Fms, discoidin domain receptor platelet-derived growth factor receptor–β, and Eph receptor kinases. Furthermore, treatment with nitrosoureas decreases Src protein in vitro,[9] and synergy was demonstrated in dasatinib with chemotherapy. A study of dasatinib as a single agent in recurrent glioblastoma suggested only minor benefit.[10] However, it has been hypothesized that a targeted therapy against Src kinase may increase the antitumor effect of cyclonexyl-chloroethyl-nitrosourea (CCNU). We report the results of the safety cohort of a European Organization for Research and Treatment of Cancer (EORTC) phase I/II trial of dasatinib in combination with CCNU in recurrent glioblastoma patients.

Patients and Methods

Eligibility Criteria

Patients eligible for this study had histologically verified glioblastoma according to the WHO 2007 classification, recurrent after standard RT with concomitant and adjuvant temozolomide. Prior exposure to other agents as part of initial therapy was allowed if at least 4 weeks had elapsed since the end of treatment and if patients had recovered from all toxicity. Other than reoperation, no prior therapy for recurrent disease was allowed. In case of reoperation, a postsurgery MRI within 48 h after surgery must have been available, but presence of residual disease was not required because the primary endpoint for the phase II part of the study was progression-free survival (PFS) at 6 months (PFS6). In patients without residual disease or with nonmeasurable disease after a second surgery, PFS was calculated from enrollment to disease progression at MRI. For non-operated patients, documentation was needed (within 2 weeks prior to enrollment) of at least 1 bidimensionally contrast-enhancing lesion (diameter $\geq$2 cm).

In order to exclude pseudoprogression[11–13] or radiation necrosis, RT must have been completed at least 3 months prior to enrollment. Patients having received prior high-dose RT ($\geq$65 Gy) were not eligible unless the recurrence was histologically confirmed. Other inclusion criteria were age $\geq$18 years, WHO performance status 0–2, stable or decreasing dose of corticosteroids for at least 1 week prior to treatment start, neutrophils $\geq 1.5 \times 10^9$ cells/L, platelets $\geq 100 \times 10^9$ cells/L, bilirubin $<1.5 \times$ upper limit of the normal range (ULN), alkaline phosphatase and transaminases $<2.5 \times$ ULN, and serum creatinine $<1.5 \times$ ULN. Patients requiring anticonvulsant therapy must have been switched to a non-enzyme-inducing antiepileptic drug (non-EIAED) $\geq$2 weeks prior to study entry. Adequate pulmonary function was required, including diffusion lung capacity of carbon monoxide (DLCO) $\geq$60% of the predicted value. Clinically normal cardiac function was required, without cardiac insufficiency (New York Heart Association grades III and IV), unstable angina, or arrhythmia. Patients with ischemic heart disease in the prior 12 months must have been stable (eg, prior angina under appropriate therapy). Written informed consent from all patients and approval from the institutional review boards of participating centers were obtained.

Study Design

EORTC 26083/BMS CA180-274 (EudraCT [European Union Drug Regulating Authorities Clinical Trial] no. 2009-010576-21, NCT 00948389) was initially designed as an open-label, multicenter, randomized, phase II study of CCNU alone or combined with dasatinib in patients with recurrent glioblastoma, with an initial dose-escalating "run-in" phase to establish the safety of this combination. This initial dose-escalating phase required expansion and is therefore considered a phase I part to establish the recommended dosing regimen of the combination.

Dose-Escalation Procedure

CCNU was administered orally at a dosage of 110 mg/m^2 every 6 weeks up to a maximum of 6 cycles or until progression, unacceptable toxicity, or death; a cycle was defined as 6 weeks. Dose reduction to 90 mg/m^2 was permitted. Dose interruption was also permitted.

For the initial 3 patients, cycle 1 was dasatinib at 100 mg once daily (q.d.) escalated to 100 mg b.i.d. in cycle 2. In the absence of dose-limiting toxicity (DLT) in the first 3 patients, the 7 subsequent patients were to be treated at 100 mg b.i.d. In case of 1 DLT, 3 additional patients would be included. If no DLT was observed in these 3 additional patients, the 4 subsequent patients would be included starting at 100 mg b.i.d. In case of 2 DLTs in up to 6 patients, an intermediate dose would be explored. The recommended dose from the safety part was defined as the dose of dasatinib that leads to DLT in not more than 2 out of 10 patients. DLT was defined as an adverse drug reaction (according to Common Terminology Criteria for Adverse Events v3.0) as follows: absolute neutrophil counts $<0.5 \times 10^9$/L lasting for 7 consecutive days, febrile neutropenia, thrombocytopenia grade 4, any grades 3–4 nonhematological toxicity (except nausea, vomiting, or fever controlled with appropriate measures), or any toxicity that prevented administration of at least 70% of the intended dose of either agent. DLTs were documented over the first 2 cycles (12 weeks) for patients starting with dasatinib 100 mg q.d. and escalated to dasatinib 100 mg b.i.d. and only in cycle 1 for patients starting with dasatinib 100 mg b.i.d. Patients discontinuing study during the DLT observation period for any reason other than toxicity were replaced.

Drug Administration

Both drugs are available as oral formulations; treatment was given in an outpatient setting. Patients were instructed to take dasatinib with the suggestion that the daily intake of study drug be at approximately the same time each day. The tablets were not to be chewed or crushed and were to be swallowed whole with water. Vomited or missed doses were not replaced. Patients were instructed to not drink pomegranate or grapefruit juice during the study.

Pretreatment Evaluation

We obtained at baseline medical history; performance status; physical examination, including neurological evaluation (Medical Research Council neurological function scale); DLCO test for lung function; status of corticosteroid administration; and a contrast-enhanced brain MRI (within 2 weeks prior to registration/randomization) and ECG. Complete blood counts and serum chemistry were also obtained at baseline and every 2 weeks. Disease assessment was performed every 6 weeks ($\pm$ 1 week) or sooner, if clinically indicated during the study, by contrast-enhanced brain MRI. Response evaluation was based on neuroradiological MRI, neurological conditions, and corticosteroid doses according to the Macdonald criteria.[14] A DLCO test for lung function was obtained every 12 weeks.

Pharmacokinetic Evaluation

Pharmacokinetic evaluations were conducted to study the plasma profile of dasatinib. Serial blood samples were collected on days 1 (with CCNU) and 8 (without CCNU) of cycles 1 and 2 before dasatinib administration and at 1 h, 2 h, 4 h, 6 h, 8 h, and 24 h after administration of dasatinib. The complete pharmacokinetic sampling was obtained in both cycle 1 and cycle 2 and optionally in case of a dose reduction.

Histological Diagnosis and Molecular Markers

Central pathology review was performed according to the WHO 2007 classification criteria on full sections stained with hematoxylin and eosin. Available for most cases were glial fibrillary acidic protein, MAP-2, MIB-1, and reticulin silver stain. All samples were screened for the most common mutation in the isocitrate dehydrogenase (IDH)–1 gene R132H by immunohistochemistry using the specific antibody mIDH1R132H (clone H14)[15] (Dianova). The methylation status of the promoter of the O^6-methylguanine-DNA methyltransferase (MGMT) gene was tested by MDxHealth, using quantitative methylation-specific PCR.[16]

Statistical Analysis

All analyses, except pharmacokinetics, are descriptive. Baseline characteristics, exposure to treatment, drug safety profile, and further anticancer treatment were described in the safety population, defined as "all enrolled patients who have received at least 1 dose of study medication." Accrual statistics were computed for patients in the enrolled population, defined as "all enrolled patients, whether treated or not." Maximum tolerated dosage (MTD) and recommended dose were identified among all patients included in the evaluable population, defined as "all enrolled patients who completed the period for DLT observation." Differences

Table 1. Baseline demographics and patient characteristics

	Total ($N = 26$) *n* (%)
Sex	
Male	20 (76.9)
Female	6 (23.1)
Age	
Median	54.8
Range	38.8–70.7
Histological diagnosis of primary disease (by local pathologist)	
Glioblastoma multiforme	22 (84.6)
Glioblastoma with oligodendroglial component	4 (15.4)
For non-operated patients: presence of at least 1 target lesion	19 (73.1)
For operated patients: presence of residual disease	
No	1 (3.8)
Yes, nontarget lesion(s)	1 (3.8)
Yes, target lesion(s)	5 (19.2)
Karnofsky performance status	
0	14 (53.8)
1	10 (38.5)
2	2 (7.7)
Surgery for recurrence	
No	19 (73.1)
Yes	7 (26.9)
Corticosteroids	
No (or physiologic replacement only)	17 (65.4)
Yes, stable or decreasing dose for at least 1 week	9 (34.6)
Antiepileptic drugs	
No	13 (50.0)
Yes, non-EIAED only	11 (42.3)
Yes, EIAED switched to non-EIAED	2 (7.7)
Time since surgery for primary disease (mo)	
Median	11.0
Range	4.8–62.7
Time since initial diagnosis (mo)	
Median	11.0
Range	4.8–62.7
Time since last temozolomide administration (mo)	
Median	1.9
Range	0.9–47.4
Time from last RT to treatment start (mo)	
Median	8.4
Range	2.0–60.3

between dasatinib pharmacokinetic parameters on day 1 and day 8 of cycle 1 were tested with a 2-tailed Wilcoxon exact rank test; $P < .05$ was considered significant. Disease response was assessed for patients with measurable disease, while PFS and survival were evaluated in the entire population. An analysis excluding patients who potentially received bevacizumab as part of postsurgical treatment was performed to evaluate response rate, PFS, and survival in this subgroup.

Results

Between October 2009 and February 2011, 28 patients were screened for study participation at 5 treatment centers. Two patients were not considered eligible and were not enrolled owing to low DLCO. Three patients received postsurgical biotherapy (bevacizumab or placebo in the context of the BO21990 AVAglio trial) concurrent with temozolomide and RT. Seven patients underwent a second surgery before study entry. One patient showed nonmeasurable disease and 1 no residual disease after a second surgery. Twenty-six patients started therapy and were reported as the safety population. Three patients did not complete the first cycle for reasons other than toxicity. Thus 23 patients were evaluable for determination of DLT. Patient characteristics are summarized in Table 1. Central pathology review was performed in all 26 patients enrolled in the study. All cases were considered assessable, with sufficient viable tissue and sufficient quality of the sections. One tumor (3%) did not fulfill the criteria for glioblastoma and was considered a low-grade astrocytoma. The remaining 25 tumors were diagnosed as glioblastoma, of which 2 (8%) were subtyped as gliosarcoma. One glioblastoma exhibited an IDH1 mutation. The *MGMT* promoter was classified as methylated in 8 and unmethylated in 12 cases (invalid in 6, owing to insufficient quantity or quality of DNA). The molecular markers suggest no selection for good prognostic factors in the study population.

Sequence of Dose Levels, Tolerance, and DLTs

Five dose levels (DLs) were explored (Table 2) and a total of 42 cycles administered. The majority of patients (21/26) received only 1 treatment cycle; the maximum number of cycles was 8. Reasons for treatment discontinuation were progression of disease or death in 22 (84.6%), grade 4 thrombocytopenia in 3 (11.5%), and both disease progression and toxicity in 1 (3.8%). Median CCNU dose intensity was 90.2% (range: 64.7–104.5) of the calculated theoretical dose, while median dasatinib dose intensity was 70.3% (range: 51.7–100.0).

DLTs, mainly myelosuppression, occurred in 10 patients. As severe myelosuppression was demonstrated in 3/5 patients at DL-1A (CCNU 110 mg/m^2/cycle and dasatinib 100 mg q.d. at cycle 1 and 100 mg b.i.d. at cycle 2), the CCNU dose was subsequently reduced to DL-1B (CCNU 90 mg/m^2/cycle and dasatinib 100 mg q.d. at cycle 1 and 100 mg b.i.d. at cycle 2). None of the 3 patients in DL-1B experienced DLT. Thus dasatinib was subsequently escalated per protocol to 100 mg b.i.d. in DL-2 (CCNU 90 mg/m^2/cycle and dasatinib 100 mg b.i.d.). At this DL, dose-limiting grade 4 thrombocytopenia was observed in 1 patient, and in 2 patients dasatinib was interrupted owing to grade 2 and grade 3 thrombocytopenia, respectively. We explored an intermediate DL-3A (CCNU 90 mg/m^2/cycle and dasatinib 150 mg/d: 100 mg AM and 50 mg PM). Dose-limiting thrombocytopenia was observed in 4/9 patients treated with DL-3A: 3 patients had grade 4 thrombocytopenia (with grade 4 neutropenia in 1 patient), and 1 patient had grade 3 thrombocytopenia. The subsequent further dose was reduced to DL-3B (CCNU 90 mg/m^2/cycle and dasatinib 100 mg q.d.), which was well tolerated with no significant toxicities in the patient treated. Therefore, dasatinib 100 mg q.d. in combination with CCNU 90 mg/m^2 every 6 weeks does qualify as the MTD to be explored in further trials. Other toxicities observed are summarized in Tables 3 and 4.

Overall, dasatinib in combination with CCNU incurs substantial hematological toxicity, with grade 3 or 4 thrombocytopenia and neutropenia in 11 patients (42.3%) and 7 patients (26.9%), respectively. Twelve patients experienced at least 1 grade 3 toxicity and 4 nonhematological toxicities. Fourteen patients (53.8%) experienced serious adverse events. Pleural effusion, potentially a specifically dasatinib-related toxicity, was observed in 1 patient after 4 cycles.

Pharmacokinetics

Of the 16 patients were studied for pharmacokinetics, 15 had sufficient data to estimate all pharmacokinetic

Table 2. Number of patients per dose level

	CCNU	Dasatinib	Number of Patients (%)	Not Evaluable for DLTs*
Dose Level (DL)				
DL-1A	110 mg/m^2/cycle	100 mg q.d. at cycle 1 and 100 mg b.i.d. at cycle 2	6 (23.1)	Pt 3
DL-1B	90 mg/m^2/cycle	100 mg q.d. at cycle 1 and 100 mg b.i.d. at cycle 2	3 (11.5)	
DL-2	90 mg/m^2/cycle	100 mg b.i.d.	7 (26.9)	Pt 11 and 17
DL-3A	90 mg/m^2/cycle	150 mg/day (100 mg AM and 50 mg PM)	9 (34.6)	
DL-3B	90 mg/m^2/cycle	100 mg q.d.	1 (3.8)	
Total			26 (100.0)	

Abbreviations: CCNU, cyclonexyl-chloroethyl-nitrosourea; DTL, dose-limiting toxicity; q.d., once daily.

Table 3. Number of patients per dose level (DL) and grades 3/4 hematological toxicity

	Dose Level					Total ($N = 26$)
	DL-1A ($n = 6$) n (%)	DL-1B ($n = 3$) n (%)	DL-2 ($n = 7$) n (%)	DL-3A ($n = 9$) n (%)	DL-3B ($n = 1$) n (%)	n (%)
At least one grade 3 or 4	5 (83.3)	1 (33.3)	4 (57.1)	8 (88.9)	0 (0.0)	18 (69.2)
Neutropenia	2 (33.3)	1 (33.3)	0 (0.0)	4 (44.4)	0 (0.0)	7 (26.9)
Leukopenia	1 (16.7)	1 (33.3)	0 (0.0)	4 (44.4)	0 (0.0)	6 (23.1)
Lymphocytopenia	4 (66.7)	0 (0.0)	1 (14.3)	5 (55.6)	0 (0.0)	10 (38.5)
Thrombocytopenia	4 (66.7)	0 (0.0)	2 (28.6)	5 (55.6)	0 (0.0)	11 (42.3)
Anemia	0 (0.0)	0 (0.0)	1 (14.3)	0 (0.0)	0 (0.0)	1 (3.8)
At least one grade 3 or 4	0 (0.0)	0 (0.0)	0 (0.0)	1 (11.1)	0 (0.0)	1 (3.8)
Hypokalemia	0 (0.0)	0 (0.0)	0 (0.0)	1 (11.1)	0 (0.0)	1 (3.8)

Table 4. Number of patients per dose level (DL) and grades ≥ 3 nonhematological toxicities

	DL-1A ($n = 6$) Grade			DL-1B ($n = 3$) Grade			DL-2 ($n = 7$) Grade			DL-3A ($n = 9$) Grade			DL-3B ($n = 1$) Grade			Total ($N = 26$) Grade		
	3	4	5	3	4	5	3	4	5	3	4	5	3	4	5	3	4	5
Event																		
Fatigue							1									1		
Anorexia							1									1		
Vomiting										1						1		
Hemorrhage		1															1	
Neuropathy: motor	1						1			2						4		
Seizure							1			1	1					2	1	
Speech impairment				1			1			1						3		
Other: central motor weakness							2									2		
Infection: normal absolute neutrophil count							1									1		
Pain: musculoskeletal–muscle										1						1		
Pain: headache											1						1	
Other toxicities									1									1

Table 5. Cycle 1 dasatinib pharmacokinetics after administration of 100 mg dasatinib and CCNU (C1D1) or 100 mg dasatinib alone (C1D8).

Day	n	C_{max}, ng/mL (SD)	T_{max}, h (SD)	$T_{\frac{1}{2}}$, h (SD)	Cl/F, L/h (SD)	Vd/F, L (SD)
C1D1	15	124 (84)	1.3 (0.5)	3.49 (1.70)	263 (100)	1213 (456)
C1D8	15	131 (76)	1.7 (1.8)	3.68 (1.90)	352 (381)	2101 (3011)
C2D1	2	78.5 (−)	2.0 (−)	2.72 (−)	364 (−)	1409 (−)
C2D8	2	162 (−)	1.5 (−)	2.45 (−)	231 (−)	878 (−)
	P	.762	1.00	.463	.820	.843

Differences between dasatinib pharmacokinetic parameters on day 1 and day 8 of cycle 1 were tested with a 2-tailed Wilcoxon exact rank test, where $P < .05$ was considered significant.
Abbreviations: CCNU, cyclonexyl-chloroethyl-nitrosourea; SD: standard deviation.

parameters on both days 1 and 8 of cycle 1. Cycle 2 pharmacokinetics were available in 2 patients. There was no statistically significant difference in dasatinib pharmacokinetics between day 1 (with CCNU) and day 8 (dasatinib alone) (see Table 5 and Fig. 1), suggesting the absence of a significant effect of CCNU coadministration on dasatinib pharmacokinetic results.

Antitumor Activity

Response was evaluable in 24/26 patients—after a second surgery, 1 patient had no residual disease, and 1 patient had nonmeasurable disease. One partial response (4%) and 6 disease stabilizations (25%) were achieved.

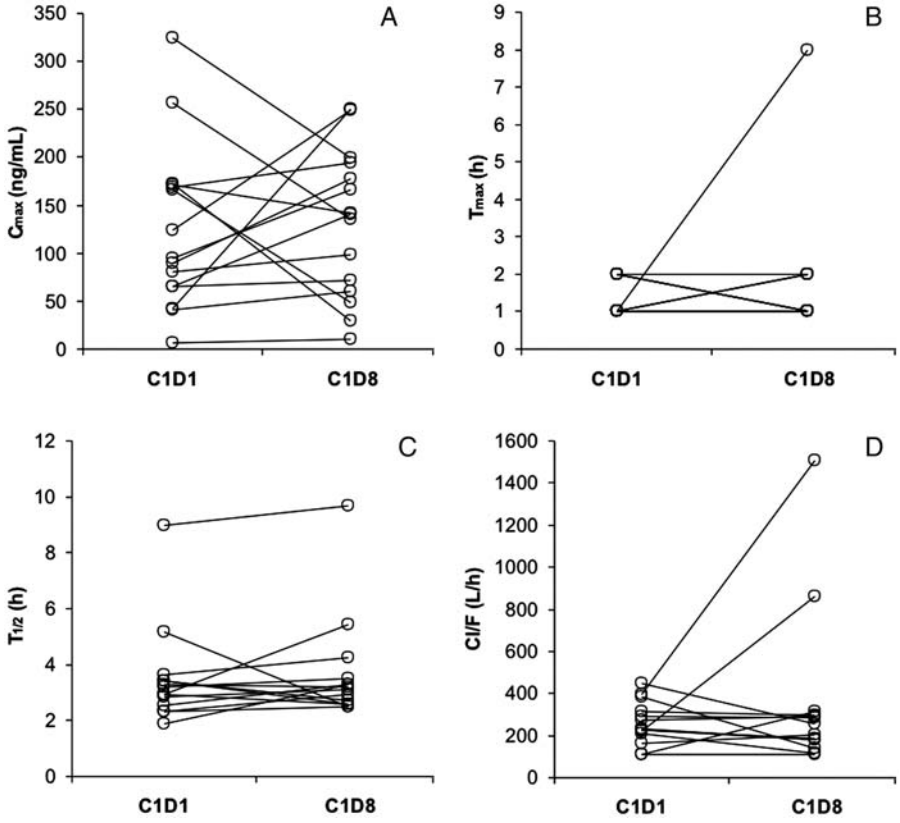

Fig. 1. C_{max} (A, $n = 16$, $P = .762$), T_{max} (B, $n = 16$, $P = 1.00$), half-life (C, $n = 15$, $P = .462$), and apparent clearance (D, $n = 15$, $P = .820$) of dasatinib, in the presence (C1D1) and absence (C1D8) of CCNU. C_{max}, maximum plasma concentration of drug. T_{max}, time that drug is present at maximum concentration.

In the entire population, median PFS was 1.35 months (95% confidence interval [CI]: 1.2–1.4), PFS6 was 7.7%, and median survival was 6.4 months (95% CI: 3.8–10.2).

The MTD was DL-3A (CCNU 90 mg/m^2/cycle and dasatinib 150 mg/d), and at this dosage PFS6 was 0%. *MGMT* methylation status (as determined from the initial diagnostic tissue; tissue at recurrence was not available) did not significantly correlate with PFS or survival. However, these numbers were too small to draw any conclusion.

Since 3 patients may have been enrolled for bevacizumab refractory disease, which could have negatively biased the results, the analysis was also performed excluding these 3 patients. In this group, 1 partial response (4%) and 6 disease stabilizations (26%) were achieved. Median PFS was 1.35 months (95% CI: 1.3–1.5), PFS6 was 8.7% (95% CI: 1.5–24.2), and median survival was 8.25 months (95% CI: 3.8–12.2).

Discussion

At recurrence after first-line adjuvant chemoradiotherapy, chemotherapy options are limited, with no standard treatment available. In the United States, the FDA approved single-agent bevacizumab based on phase II data.[4,17] However, in the European Union, the European Medicines Agency did not approve this agent, and nitrosoureas are considered the standard of care. Many promising targeted new therapies have been investigated in the treatment of glioblastoma.[2,3,18–24] Src kinases, which are involved in cell motility and cellular invasion, can be important drivers in gliomagenesis because invasion and infiltration are hallmarks of these tumors.

We started this phase I/II trial with the aim to combine the inhibition of Src-dependent pathways with nitrosourea chemotherapy. Dasatinib is an established agent in chronic myeloid leukemia and Philadelphia-chromosome+ acute lymphoblastic leukemia. The most frequently reported toxicities are diarrhea, rash, edema, fatigue, bleeding, pleural effusion, dyspnea, nausea and vomiting, pyrexia, and hematotoxicity. In hematological malignancies, a dosage of 100 mg b.i.d. was found safe and efficacious, whereas in patients with solid tumors this dosage appeared to be associated with significant toxicity (gastrointestinal intolerance, fatigue, headache, musculoskeletal pain, and pleural and pericardial effusions). In the present trial, CCNU at the standard dose of 110 mg/m^2 in combination with dose-escalating dasatinib resulted in significant toxicity, namely thrombocytopenia. Despite the fact that the reason for treatment discontinuation was progression of disease or death in 22 patients (84.6%), grade 4 thrombocytopenia led to treatment interruption in 3

patients (11.5%), and both disease progression and toxicity were responsible for discontinuation in 1 patient (3.8%). Grade 3 or 4 neutropenia was recorded in about 25% of patients and grade 3 or 4 thrombocytopenia in about 40% of patients. Therefore, substantial dose reductions of both agents were necessary, leading to potentially insufficient dose intensity. Pharmacokinetic evaluation did not show any significant interaction with CCNU to explain the relevant toxicity that we found across many DLs in this study, and therefore a synergistic effect on the toxicity profile is possible. Nevertheless, the overlapping toxic effects resulted in suboptimal exposure to both agents, since the MTD (DL-3A) was found with CCNU at the dose of 90 mg/m^2/cycle and dasatinib at 150 mg/day.

A recent phase II trial by the Radiation Therapy Oncology Group (RTOG 0627) evaluated single-agent dasatinib. At the dosage of 100 mg b.i.d., dasatinib was well tolerated; only 2/26 patients (7.7%) experienced grade 3 neutropenia. However, no clinically significant activity was reported (PFS6 $<$10% with no responses).[10] Similarly, a recent phase I trial with dasatinib in combination with the anti–epidermal growth factor receptor nonmyelotoxic agent erlotinib did allow higher exposure to dasatinib, but no activity of the combination was found, PFS6 being 2% and without evidence of radiological responses.[25]

In our trial, 81% of patients received only 1 cycle of therapy owing to tumor progression in 69% of patients and grade 4 toxicity in 12% of patients. Only 2/26 treated patients remained on study more than 6 months. Although the treatment was not reasonably tested for efficacy, the response rate (4%) and PFS-6 (8%), lower than in historical control, together with the hematological toxicity profile that limited the drug exposure of both CCNU and dasatinib lead to trial closure before the planned phase II part of the study.

Funding

Bristol-Myers Squibb is the legal sponsor of this study.

Acknowledgment

We thank the EORTC Headquarters for their contributions to the study's operations.

Conflict of interest statement. L. C. S. receives salary and stock as an employee of Bristol-Myers Squibb. Advisory affiliations—R.S.: Merck & Co, MSD, Celgene, and Roche; M.H.: Merck & Co and MSD. Paid consulting—M. H.: MDxHealth and Merck-Serono; M.J.v.d.B.: Bristol-Myers Squibb; F. L.-D.: Bristol-Myers Squibb.

References

1. Stupp R, Mason WP, van den Bent MJ, et al. Radiotherapy plus concomitant and adjuvant temozolomide for glioblastoma. *N Engl J Med*. 2005;352(10):987–996.
2. van den Bent MJ, Brandes AA, Rampling R, et al. Randomized phase II trial of erlotinib versus temozolomide or carmustine in recurrent glioblastoma: EORTC Brain Tumor Group study 26034. *J Clin Oncol*. 2009;27(8):1268–1274.
3. Wick W, Puduvalli VK, Chamberlain MC, et al. Phase III study of enzastaurin compared with lomustine in the treatment of recurrent intracranial glioblastoma. *J Clin Oncol*. 2010;28(7):1168–1274.
4. Friedman HS, Prados MD, Wen PY, et al. Bevacizumab alone and in combination with irinotecan in recurrent glioblastoma. *J Clin Oncol*. 2009;27(28):4733–4740.
5. Chi AS, Wen PY. Inhibiting kinases in malignant gliomas. *Expert Opin Ther Targets*. 2007;11(4):473–496.
6. Du J, Bernasconi P, Clauser KR, et al. Bead-based profiling of tyrosine kinase phosphorylation identifies SRC as a potential target for glioblastoma therapy. *Nat Biotechnol*. 2009;27(1):77–83.
7. Angers-Loustau A, Hering R, Werbowetski TE, Kaplan DR, Del Maestro RF. SRC regulates actin dynamics and invasion of malignant glial cells in three dimensions. *Mol Cancer Res*. 2004;2(11):595–605.
8. Lund CV, Nguyen MT, Owens GC, et al. Reduced glioma infiltration in Src-deficient mice. *J Neurooncol*. 2006;78(1):19–29.
9. Faustman EM, Sweeney C. Effects of ethylnitrosourea on expression of proto-oncogene pp60c-src and high-molecular-weight neurofilament protein in rodent embryo central nervous system cells in vitro. *Toxicol Appl Pharmacol*. 1994;128(2):182–188.
10. Lassman AB, Wang M, Gilbert MR, et al. Phase II trial of dasatinib in target selected patients with recurrent glioblastoma (RTOG 0627). *Neuro-Oncology*. 2011;13:(suppl 3):iii64.
11. Brandes AA, Tosoni A, Spagnolli F, et al. Disease progression or pseudoprogression after concomitant radiochemotherapy treatment: pitfalls in neurooncology. *Neuro Oncol*. 2008;10:361–367.
12. de Wit MC, de Bruin HG, Eijkenboom W, Sillevis Smitt PA, van den Bent MJ. Immediate post-radiotherapy changes in malignant glioma can mimic tumor progression. *Neurology*. 2004;63(3):535–537.
13. Brandes AA, Franceschi E, Tosoni A, et al. MGMT promoter methylation status can predict the incidence and outcome of pseudoprogression after concomitant radiochemotherapy in newly diagnosed glioblastoma patients. *J Clin Oncol*. 2008;26(13):2192–2197.
14. Macdonald DR, Cascino TL, Schold SC, Jr., Cairncross JG. Response criteria for phase II studies of supratentorial malignant glioma. *J Clin Oncol*. 1990;8(7):1277–1280.
15. Capper D, Weissert S, Balss J, et al. Characterization of R132H mutation-specific IDH1 antibody binding in brain tumors. *Brain Pathol*. 2009;20(1):245–254.
16. Vlassenbroeck I, Califice S, Diserens AC, et al. Validation of real-time methylation-specific PCR to determine O6-methylguanine-DNA methyltransferase gene promoter methylation in glioma. *J Mol Diagn*. 2008;10(4):332–337.
17. Wick W, Weller M, van den Bent M, Stupp R. Bevacizumab and recurrent malignant gliomas: a European perspective. *J Clin Oncol*. 2010;28(12):e188–189; author reply e190–182.

18. Stupp R, Hegi ME, Gilbert MR, Chakravarti A. Chemoradiotherapy in malignant glioma: standard of care and future directions. *J Clin Oncol*. 2007;25(26):4127–4136.
19. Galanis E, Buckner JC, Maurer MJ, et al. Phase II trial of temsirolimus (CCI-779) in recurrent glioblastoma multiforme: a North Central Cancer Treatment Group study. *J Clin Oncol*. 2005;23(23): 5294–5304.
20. Reardon DA, Dresemann G, Taillibert S, et al. Multicentre phase II studies evaluating imatinib plus hydroxyurea in patients with progressive glioblastoma. *Br J Cancer*. 2009;101(12):1995–2004.
21. Brandes AA, Stupp R, Hau P, et al. EORTC study 26041–22041: phase I/II study on concomitant and adjuvant temozolomide (TMZ) and radiotherapy (RT) with PTK787/ZK222584 (PTK/ZK) in newly diagnosed glioblastoma. *Eur J Cancer*. 2010;46(2): 348–354.
22. Franceschi E, Cavallo G, Lonardi S, et al. Gefitinib in patients with progressive high-grade gliomas: a multicentre phase II study by Gruppo Italiano Cooperativo di Neuro-Oncologia (GICNO). *Br J Cancer*. 2007;96(7):1047–1051.
23. Stupp R, Tosoni A, Bromberg JE, et al. Sagopilone (ZK-EPO, ZK 219477) for recurrent glioblastoma. A phase II multicenter trial by the European Organisation for Research and Treatment of Cancer (EORTC) Brain Tumor Group. *Ann Oncol*. 2011;22(9):2144–2149.
24. Raymond E, Brandes AA, Dittrich C, et al. Phase II study of imatinib in patients with recurrent gliomas of various histologies: a European Organisation for Research and Treatment of Cancer Brain Tumor Group Study. *J Clin Oncol*. 2008;26(28):4659–4665.
25. Reardon DA, Vredenburgh JJ, Desjardins A, et al. Phase 1 trial of dasatinib plus erlotinib in adults with recurrent malignant glioma. *J Neurooncol*. 2012;108(3):499–506.