Role of temozolomide after radiotherapy for newly diagnosed diffuse brainstem glioma in children: Results of a multiinstitutional study (SJHG-98)

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Role of Temozolomide after Radiotherapy for Newly Diagnosed Diffuse Brainstem Glioma in Children

Results of a Multiinstitutional Study (SJHG-98)

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BACKGROUND. The role of chemotherapy in the treatment of children with newly diagnosed diffuse brainstem glioma is uncertain. In the current study, the authors tested the efficacy of temozolomide treatment after radiotherapy (RT) in this setting.

METHODS. Patients ages 3–21 years were eligible for the current multiinstitutional study. An optional window therapy regimen consisting of 2 cycles of intravenous irinotecan (10 doses of 20 mg/m^2 per day separated by 2 days of rest per cycle) was delivered over 6 weeks and was followed by conventionally fractionated RT. The 5-day schedule of temozolomide (200 mg/m^2 per day) was initiated 4 weeks after RT and was continued for a total of 6 cycles. The pharmacokinetics of temozolomide and its active metabolite, 5-(3-methyltriazen-1-yl)imidazole-4-carboxamide (MTIC), were analyzed during Cycles 1 and 3.

RESULTS. Thirty-three patients (median age at diagnosis, 6.4 years) were enrolled. Of the 16 patients who received window therapy, 6 had irinotecan treatment discontinued due to clinical progression ($n = 5$) or toxicity ($n = 1$); the remaining 10 experienced disease stabilization after 2 cycles. All patients completed RT (median dose, 55.8 gray). Twenty-nine patients received a combined total of 125 cycles of temozolomide. Grade 3/4 neutropenia and thrombocytopenia occurred in 33% and 29% of all temozolomide cycles, respectively. In approximately one-third of the cycles, dose reduction was required due to myelosuppression. No correlation was demonstrated between temozolomide/MTIC exposure and myelosuppression at the conclusion of Cycle 1. All patients died of disease progression (median survival, 12 months). The estimated 1-year survival rate was 48% (standard error, 8%).

CONCLUSIONS. The administration of temozolomide after RT did not alter the poor prognosis associated with newly diagnosed diffuse brainstem glioma in children. ***Cancer* 2005;103:133–9.** © *2004 American Cancer Society.*

KEYWORDS: diffuse brainstem glioma, temozolomide, children, irinotecan.

The prognosis for children with newly diagnosed diffuse brainstem glioma is very poor. Conventionally fractionated local radiotherapy (RT) remains the standard of care, leading to temporary clinical improvement in a substantial percentage of patients.[1]

The role of chemotherapy in the treatment of children with diffuse brainstem glioma is not well defined. Multiple trials involving various regimens administered in conjunction with RT have not demonstrated any improvement in outcome.[1] Clinical trials for patients with diffuse brainstem glioma continue to test the efficacy of novel agents administered concurrently with or subsequent to RT.

Temozolomide is an orally administered methylating agent that

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has shown promising responses in a subset of adults with recurrent or newly diagnosed high-grade glioma.[2–6] Two Phase I studies characterized the toxicity profile and maximum tolerated dose of temozolomide when administered on a 5-day schedule to children.[7,8] Like adult trials, these pediatric studies showed that myelosuppression was the main side effect of temozolomide. In one of these studies, 2 of 5 children with recurrent high-grade astrocytoma and 1 of 10 children with diffuse brainstem glioma experienced objective responses to treatment.[7] In the other study, one patient with high-grade astrocytoma had a partial response.[8]

On the basis of the results observed in adults and the early responses documented in pediatric trials, we conducted a multiinstitutional study to evaluate the role of temozolomide in the treatment of newly diagnosed infiltrative glial neoplasms in children. Eligible patients were divided into two treatment groups. One group consisted of patients with diffuse brainstem glioma, and the other consisted of patients with other high-grade gliomas arising outside the brainstem. We report the results of this treatment approach as well as the pharmacokinetic behavior of temozolomide and its active metabolite, 5-(3-methyltriazen-1-yl)imidazole-4-carboximide (MTIC), in children and adolescents with diffuse brainstem glioma.

MATERIALS AND METHODS

Eligibility Criteria

Patients ages 3–21 years with newly diagnosed diffuse brainstem glioma were enrolled in the current study (SJHG-98) by 3 participating institutions (Children's Medical Center, Dallas, TX; St. Jude Children's Research Hospital, Memphis, TN; and Texas Children's Hospital, Houston, TX). Histologic confirmation was not necessary for any patient in whom typical magnetic resonance imaging (MRI) scans revealed a pontine-based intrinsic infiltrative lesion exerting a mass effect on contiguous structures. No previous therapy, excluding corticosteroid treatment, was permitted.

Adequate bone marrow (hemoglobin concentration $>$ 8 g/dL, leukocyte count $>$ 2500/mm^3, and platelet count $>$ 100,000/mm^3), hepatic (liver function values $<$ 5 times the normal laboratory levels and bilirubin concentration $\leq$ 2.5 mg/dL), and renal (serum creatinine concentration $\leq$ 2 mg/dL) functioning was required at study entry. Females of childbearing age were required not to be pregnant for inclusion in the current study.

Treatment was planned to begin as soon as possible (i.e., once written informed consent was obtained) within 28 days of diagnosis. The study was approved by the institutional review board at each participating institution.

Treatment Plan

Children with diffuse brainstem glioma were eligible to receive optional upfront window therapy that consisted of 2 cycles of irinotecan (Pharmacia & Upjohn, Kalamazoo, MI) administered at 3-week intervals. Irinotecan was administered intravenously at a daily dose of 20 mg/m^2 over 5 consecutive days, after which there was a 2-day rest period and then 5 more consecutive days of administration.

Conventionally fractionated 3-dimensional conformal RT using a megavoltage accelerator began within 2 weeks of the completion of irinotecan therapy for patients receiving the window therapy regimen. The recommended total dose was 55.8 gray (Gy).

Temozolomide was supplied in the form of either 20 or 100 mg capsules by the Schering-Plough Research Institute (Kenilworth, NJ). The 5-day schedule of temozolomide therapy (200 mg/m^2 per day) began 4 weeks after the end of RT and continued for a total of 6 cycles. Patients were asked to fast for 2 hours before and 1 hour after temozolomide administration. Initially, the minimum interval between cycles was defined as 21 days. This interval was later modified to 28 days because of the prolonged hematopoietic recovery time required by patients. The criteria for initiating subsequent cycles of temozolomide included an absolute neutrophil count $\geq$ 1000/mm^3, an unsupported platelet count $\geq$ 50,000/mm^3, a hemoglobin concentration $\geq$ 8 g/dL, and full recovery from all nonhematopoietic toxicity. A dosage reduction of 20% was recommended for treatment delays that exceeded 7 days secondary to myelosuppression. Further dosage reduction in subsequent cycles was allowed if prolonged hematopoietic toxicity recurred. Treatment side effects were graded according to the National Cancer Institute Common Toxicity Criteria (Version 2.0).

Supportive Care

Intravenous atropine at a dose of 0.01 mg/kg (maximum total dose, 0.4 mg) was recommended for symptoms of acute cholinergic syndrome associated with irinotecan use. Loperamide treatment was initiated at the first occurrence of poorly formed or loose stools or at the first indication of increased frequency of bowel movements after the start of irinotecan therapy. The loperamide dose was titrated in response to each patient's symptoms.

Irradiated, leukocyte-depleted blood products were used to maintain platelet counts $>$ 20,000–30,000/mm^3 and hematocrit values $>$ 20–25%. All pa-

tients received *Pneumocystis cariini* prophylaxis beginning with the first cycle of temozolomide therapy. Antiemetic drugs were used at the discretion of each investigator during window therapy and RT, but ondansetron use was strongly encouraged during the administration of temozolomide.

Episodes of fever and neutropenia were managed according to institutional guidelines. A single-lumen Hickman catheter (C.R. Bard, Inc., Salt Lake City, UT) was recommended for all patients for whom central venous access was required. Administration of granulocyte colony-stimulating factor after the completion of each cycle of temozolomide therapy was not routinely recommended.

Radiologic Evaluation

Objective radiologic responses were ascertained by comparing consecutive brain MRIs performed at regular intervals. Radiologic findings were not centrally reviewed.

A complete response (CR) entailed the disappearance of all measurable lesions. A partial response (PR) was defined as a reduction of $\geq 50\%$ in the sum of the products of the maximum perpendicular diameters of all measurable lesions. Progressive disease was defined as an increase of $> 25\%$ in the sum of the products of the maximum perpendicular diameters of any lesions or the appearance of any new lesions. All other responses were classified as stable disease (SD).

Temozolomide Pharmacokinetics

We evaluated the pharmacokinetics of temozolomide and MTIC on Days 1 and 5 of the first and third cycles. Measurements were performed on Day 2 or Day 4, respectively, when Day 1 or Day 5 fell on a weekend. Some patients had samples collected during Cycle 2 instead of Cycle 3 for logistic reasons (e.g., venous access). Three milliliters of whole blood was collected in a lithium heparin tube before and at 0.25, 0.5, 1, 2, 2.5, 3, 6, and 8 hours after temozolomide administration. Samples were processed and analyzed using isocratic high-performance liquid chromatography.[9]

Temozolomide and MTIC plasma concentration–time data were modeled using a maximum a posteriori Bayesian estimation as implemented in the ADAPT II software package (University of Southern California Biomedical Simulations Resource, Los Angeles, CA). The parameter estimates used in the current study were described by Panetta et al.[9] A first-order absorption one-compartment linear model, which included first-order MTIC formation and elimination, was used to simultaneously describe the pharmacokinetics of temozolomide and MTIC.[9] The area under the concentration-time curve (AUC_{0-24}) for each compound was calculated using the log-linear trapezoidal rule.

TABLE 1
Patient Characteristics at Diagnosis ($n = 33$)

Characteristic	No. of patients (%)
Median age in yrs (range)	6.4 (3.1–15.2)
Gender	
Male	15 (45)
Female	18 (55)
Race/ethnicity	
Caucasian	24 (73)
African American	7 (21)
Hispanic	2 (6)
Treating institution	
St. Jude Children's Research Hospital (Memphis, TN)	29 (88)
Texas Children's Medical Center (Houston, TX)	2 (6)
Children's Medical Center (Dallas, TX)	2 (6)
Histology	
Not available	29 (88)
Glioblastoma multiforme	1 (3)
Anaplastic astrocytoma	2 (6)
Not diagnostic	1 (3)

Statistical Analysis

Progression-free survival (PFS) and overall survival (OS) were estimated using the method of Kaplan and Meier.[10] Standard error estimates were calculated using the method described by Peto et al.[11] PFS reflected the interval between diagnosis and clinical and/or radiologic tumor progression or death. OS was defined as the interval between diagnosis and death.

Differences in pharmacokinetic parameters between Days 1 (or 2) and 5 (or 4) of Cycles 1 and 3 were analyzed using a mixed-effect model that took into account the possible intrapatient correlation caused by multiple courses and repeated pharmacokinetic determinations during each course. The model was also used to estimate interpatient and intrapatient variability. The exact Wilcoxon rank-sum test was used to assess potential correlations between temozolomide and MTIC AUC values and the occurrence of neutropenia or thrombocytopenia (Grade $\leq$ 2 vs. Grade 3/4) after Cycle 1. Two-sided P values are reported. All analyses were performed using SAS Version 8.2 (SAS Institute Inc., Cary, NC).

RESULTS

Thirty-three patients were enrolled in the current study between March 1999 and June 2002 (Table 1). The median age of patients at diagnosis was 6.4 years (range, 3.1–15.2 years). Four patients underwent biopsy ($n = 3$) or subtotal resection ($n = 1$) before they entered the study. One patient had glioblastoma mul-

tiforme, and two had anaplastic astrocytoma. Histologic analysis was not diagnostic for one patient.

Irinotecan Window Therapy

Sixteen patients began window therapy, but only 10 completed 2 cycles of irinotecan treatment. Therapy was halted prematurely in 6 patients, due to signs of progressive brainstem involvement ($n = 5$) or Grade 4 nonhematopoietic toxicity (i.e., seizures and Grade 3 leukoencephalopathy demonstrated by brain MRI; $n = 1$). The patient who developed Grade 4 nonhematopoietic toxicity had previously experienced Grade 3 diarrhea, hyponatremia, and neutropenia.

Only 2 of 10 patients who completed upfront window therapy experienced Grade 3 or 4 toxicity. Grade 3 diarrhea developed in 1 patient, and significant myelosuppression, consisting of Grade 4 neutropenia ($n = 1$) and Grade 3 anemia ($n = 1$), was also observed.

All 10 patients who completed 2 cycles of irinotecan therapy experienced disease stabilization as indicated by radiologic studies.

Radiotherapy

All patients completed RT (median dose, 55.8 Gy; range, 50.4–57.8 Gy). Information regarding radiologic responses after RT was available for 32 patients—7 experienced PR, and 25 had SD.

Temozolomide Treatment

Four patients did not receive temozolomide, either at the parents' request ($n = 3$) or because of noncompliance with treatment ($n = 1$). Three patients began temozolomide therapy but had treatment interrupted at the parents' request (after the first cycle), because of loss to follow-up (after two cycles), or because of noncompliance (after two cycles). A total of 125 cycles of temozolomide therapy (median number of cycles per patient, 4) were administered. The toxicity associated with temozolomide use was primarily hematopoietic. Grade 3/4 neutropenia, thrombocytopenia, and anemia were documented in 33%, 29%, and 7% of all cycles, respectively. Only 1 cycle of temozolomide therapy was complicated by febrile neutropenia, and 1 patient experienced a Grade 3 rash that was attributed to the medication. In response to prolonged Grade 3/4 myelosuppression, the dose of temozolomide was reduced in a total of 45 cycles (36%) for 14 patients. The median interval between Cycles 1 and 2 was 35 days (range, 19–46 days). The median intervals between the remaining cycles of temozolomide therapy were 33, 35, 29, and 32 days, respectively.

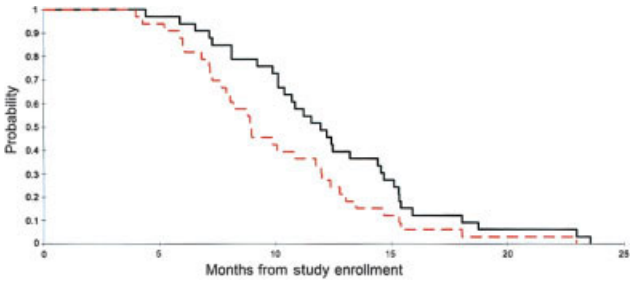

FIGURE 1. Overall (solid black line) and progression-free (dashed red line) survival probabilities for all 33 patients enrolled in the current study

Outcome

All patients experienced progression and died of disease. The median time to disease progression was 8.8 months (range, 3.9–17.9 months). The site of radiologic progression was documented in 22 patients. Twenty patients (91%) initially had local disease progression only, and 2 (9%) had isolated leptomeningeal failures. The 1- and 2-year PFS estimates were 27% (standard error, 7%) and 0%, respectively (Fig. 1). The median survival duration was 12 months, and the 1-year survival rate was 48% (standard error, 8%) (Fig. 1).

Pharmacokinetics and Pharmacodynamics of Temozolomide and MTIC

Twenty-nine patients who received temozolomide were eligible to participate in the pharmacokinetic studies. At least 1 study was completed for all 29 patients during Cycle 1, and sequential studies covering Cycles 1 and 3 were conducted for 21 patients. Pharmacokinetic evaluations were performed on Days 1 (or 2) and 5 (or 4) of Cycles 1 and 3 for 23 and 19 patients, respectively. Figure 2 shows representative concentration-versus-time curves for temozolomide and MTIC.

Table 2 summarizes pharmacokinetic parameters for all 29 eligible patients on the first day of Cycle 1. Between Days 1 (or 2) and 5 (or 4) of Cycle 1, no statistical differences were observed in terms of apparent oral temozolomide clearance ($P = 0.32$), absorption delay time ($P = 0.54$), elimination half-life of temozolomide ($P = 0.15$), temozolomide AUC ($P = 0.32$), or MTIC AUC ($P = 0.66$). The elimination half-life of temozolomide was the only parameter that was statistically different between Cycles 1 and 3 (1.6 vs. 1.9 hours; $P = 0.03$). The median temozolomide and MTIC AUC values at the 200 mg/m^2 dose level on Day 1 (or 2) of Cycle 1 were 41.3 and 1.1 μg/mL per hour, respectively.

Because of the narrow range of temozolomide

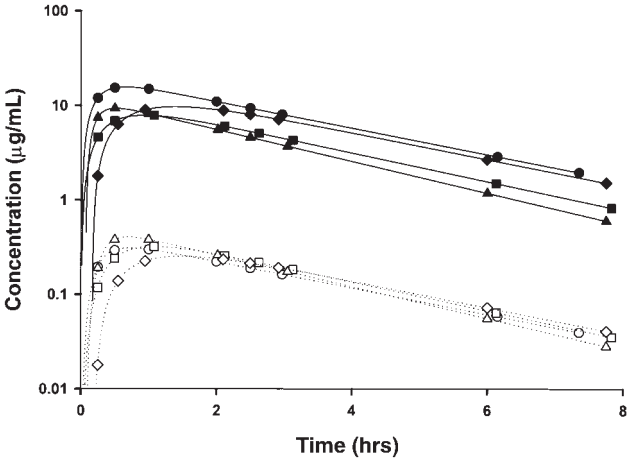

FIGURE 2. Representative temozolomide (solid line) and MTIC (dashed line) plasma concentration–time curves on Day 1 (circles) and Day 5 (squares) of Cycle 1 and on Day 1 (triangles) and Day 5 (diamonds) of Cycle 3.

TABLE 2
Summary of Pharmacokinetic Parameters in the 29 Patients Eligible To Participate in Pharmacokinetic Studies after Cycle 1

	Dose (mg/m^2)	CL_{tem} (L/m^2 per hr)	TMZ $t_{1/2}$ (hrs)	K_a (hr^{-1})	τ (hrs)
Median	200	4.50	1.76	1.58	0.04
Range	140–200	2.77–8.13	0.77–2.87	0.35–86.8	0.004–0.243

CL_{tem}: clearance of temozolomide; TMZ $t_{1/2}$: temozolomide elimination half-life; K_a: absorption rate constant; τ: absorption delay time.

doses used in the current study (140–200 mg/m^2 per day), a sigmoid dose-response model did not adequately describe the relation between drug exposure (temozolomide and MTIC) and myelosuppression. Although the median temozolomide and MTIC AUC values observed in patients who experienced Grade 3/4 neutropenia or thrombocytopenia after Cycle 1 were greater than those observed in patients who experienced Grade ≤ 2 toxicity, no statistical correlation was found between temozolomide or MTIC AUC and the extent of neutropenia ($P = 0.13$ and $P = 0.75$, respectively) or thrombocytopenia ($P = 0.29$ and $P = 0.20$, respectively).

DISCUSSION

Temozolomide generated great excitement as the first drug commercially released in the last two decades to show significant activity against a subset of high-grade gliomas in adults.[2,4,5] Despite the initial positive responses observed among children with high-grade gliomas treated in Phase I trials, we showed that the administration of temozolomide after RT does not change the poor outcome of children with newly diagnosed diffuse brainstem gliomas.

Our results corroborate the findings of a recent Phase II multiinstitutional trial that reported disappointing responses to temozolomide in children with recurrent gliomas.[12] In that study, 21 children with diffuse brainstem glioma and 34 children with other types of high-grade glioma were treated on the same 5-day schedule of temozolomide after experiencing disease recurrence or disease progression. All patients had previously received RT, and of the 23 (43%) who had previously received chemotherapy, 12 had previously received regimens containing either procarbazine or a nitrosourea. Only 1 of 18 evaluable patients with brainstem glioma had a PR to temozolomide, for an overall response rate of 6% (95% confidence interval [CI], 0–27%). Three additional patients experienced disease stabilization. Three of 25 evaluable patients with other types of high-grade glioma had PRs to treatment, for an overall response rate of 12% (95% CI, 2–31%). In the same cohort, two other children experienced temporary disease stabilization.

One other study has reported on the radiologic responses observed in six children with newly diagnosed, incompletely resected supratentorial high-grade astrocytoma who received upfront temozolomide at the same dose and according to the same schedule used in the current study.[5] A CR and SD were reported in one and two children with glioblastoma multiforme, respectively. In addition, two of three patients with anaplastic astrocytoma had SD.

The biology of diffuse brainstem gliomas could account for the low responses achieved by temozolomide therapy, but very little is known about the genetic and molecular abnormalities present in these tumors. Because the diagnosis of this neoplasm is based on typical imaging features that preclude histologic verification, thus far, only 63 tumor samples from patients with diffuse brainstem glioma have been analyzed and reported.[13–17] The presence of a mismatch repair–deficient phenotype and the expression of high levels of the O^6-methylguanine methyltranferase protein (or the lack of hypermethylation of the promoter of the corresponding gene) are two of the best known mechanisms of resistance to temozolomide, but neither has been analyzed in diffuse brainstem gliomas.

Significant myelosuppression, the primary side effect of temozolomide, was observed at a higher frequency in the current study than in most adult trials. We previously demonstrated an association between increased temozolomide clearance and both increasing age and body surface area, as well as associations between decreased temozolomide and MTIC AUC values and increasing age.[9] Although we found no corre-

lation between temozolomide and MTIC exposure and the extent of neutropenia or thrombocytopenia, we observed a trend toward higher temozolomide and MTIC AUC values during Cycle 1 in patients who experienced more severe myelosuppression. It is possible that this trend would be significant in a study involving a larger number of patients.

The results of our pharmacokinetic analysis are similar to those observed in studies of adults with advanced solid tumors who received the same temozolomide regimen at doses of 100–200 mg/m^2 per day.[18] In the current study, the apparent oral clearance of temozolomide was 4.5 L/m^2 per hour, whereas the clearance was 6.9 L/m^2 per hour in adult patients receiving equivalent dosages in the study conducted by Hammond et al.[18] . In addition, the mean temozolomide AUC reported in adults was 30 μg/mL per hour, compared with 41 μg/mL per hour for patients in the current study who were treated at the same dose. Like our study, the study of adult patients found no differences in pharmacokinetic parameters between Days 1 and 5.

Irinotecan is a novel topoisomerase I poison that has shown promising activity against xenografts of human central nervous system tumors, including high-grade gliomas derived from adults and children.[19] In clinical trials in which adults with recurrent high-grade astrocytoma were treated with different regimens of irinotecan therapy, responses were observed in as many as 15% of patients, and even larger proportions of patients experienced SD.[20–22] A smaller number of children with gliomas have also received this agent. In 2 Phase I pediatric studies, which included a total of 8 children with recurrent high-grade glioma and 3 children with recurrent brainstem glioma,[23,24] irinotecan was administered intravenously every 3 weeks as a single dose or as multiple doses given over 5 consecutive days. One patient with high-grade glioma experienced a PR, and SD was documented in five patients with high-grade glioma and one patient with brainstem glioma. In a pediatric Phase II study, irinotecan was administered weekly to 9 children with high-grade astrocytoma (newly diagnosed [$n = 4$] or recurrent [$n = 5$]) and to 5 children with progressive diffuse brainstem glioma.[25] Although four radiologic responses were observed among patients with high-grade astrocytoma, all five children with diffuse brainstem glioma experienced further disease progression. Similarly, we found no objective radiologic responses in children with newly diagnosed diffuse brainstem glioma who received this protracted irinotecan regimen.

To our knowledge, the unfavorable outcome for patients in the current study is consistent with all other reports on the treatment of diffuse brainstem glioma in children.[1] Therefore, novel therapeutic strategies for the treatment of this tumor, including the use of small-molecule inhibitors and novel drug delivery methods, continue to be investigated. Although innovative methods of drug delivery have been tested successfully in animals only,[26] several pediatric trials are already testing the efficacy or activity of novel biologic agents in children with newly diagnosed or progressive diffuse brainstem glioma. To identify additional molecules and cellular pathways that can be targeted by these novel strategies, it will be important to better understand the mechanisms that are associated with the genesis of this neoplasm. The acquisition of tumor tissue specimens for molecular analysis during treatment (when a biopsy is clinically indicated) and, particularly, at autopsy will make it possible for investigators to address these issues.

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