Phase II trial of anti-epidermal growth factor receptor radioimmunotherapy in the treatment of anaplastic astrocytoma

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Abstract

Objective The efficacy and safety of adjuvant radioimmunotherapy with ^{125}I -labeled anti-epidermal growth factor receptor 425 murine antibody (^{125}I -425 mAb) in patients with newly diagnosed anaplastic astrocytoma (AA) is evaluated in this single-institution phase II study.

Methods Patients with newly diagnosed and histologically proven AA were eligible. A total of 80 patients received three weekly intravenous injections of 1.8 GBq of ^{125}I -425 mAb following surgery and radiation therapy to a median dose of 60 Gy. Median overall survival was the primary endpoint and the secondary endpoint was toxicity.

Results The median age was 43 years (range 22–75 years), and the median Karnofsky Performance Status was 80 (range 50–100). The percentage of patients who under-

went surgical debulking, radiation therapy, temozolomide, and nitrosourea-based chemotherapy are 57.5, 99, 25, and 25 % respectively. The overall median survival was 55.6 months (95 % CI 42.7–68.5 months). The 2- and 5-year survivals were 74 and 47 %, respectively. There was no grade 3 or higher toxicity with the administration of ^{125}I -425 mAb.

Conclusion Treatment with ^{125}I -425 mAb in newly diagnosed AA patients was safe and well tolerated. Survival benefits are encouraging and warrant further study.

Keywords Radioimmunotherapy · Anaplastic · Astrocytoma · Anti-epidermal growth factor receptor · ^{125}I -labeled monoclonal antibody 425 · Glioma · Monoclonal antibody

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Introduction

Despite technological advancement in treatment and several decades of clinical and laboratory research, the prognosis of primary malignant glioma in adults remains poor. The American Cancer Society estimates that 22,910 primary malignant tumors of the central nervous system (CNS) will be diagnosed during 2012 in the USA with a similar incidence worldwide. The estimated death due to primary CNS malignancy in 2012 was 13,700 [1, 2]. The most common glial tumor in adults is glioblastoma multiforme (GBM, WHO grade IV) followed by anaplastic astrocytoma (AA, WHO grade III), and combined they account for more than 60 % of adult gliomas [3].

Among the primary brain tumors, GBM and AA have the worst prognosis with median survival of 9–12 months and 2–3 years, respectively [4]. Past studies have grouped AA with anaplastic oligodendrogliomas (AOD) and anaplastic oligoastrocytomas (AOA) despite AOD and AOA having higher chemosensitivity and better prognosis [5–7]. Historically, AA has also been grouped together with GBM in several studies. It is only recently that AA has been studied separately in prospective clinical trials [8, 9]. Standard treatment for both GBM and AA remains as surgical debulking and radiation therapy plus or minus chemotherapy. Landmark 2005 trial by Stupp et al. [10] showed survival benefit when temozolomide was added to radiation therapy in GBM patients. Since then, temozolomide has become the standard of care for GBM. However, the benefit of temozolomide in the treatment of newly diagnosed AA has yet to be proven and is still under investigation [11, 12]. The role of nitrosourea-based chemotherapy in the treatment of AA also remains controversial.

Recurrences occur at or adjacent to the site of origin despite aggressive treatment [13, 14] indicating inadequate eradication of the tumor growth. Even with new technological advances in surgery, radiation oncology, and radiology, survival has not improved in these patients. Thus, investigations into novel adjuvant treatments are highly warranted.

The epidermal growth factor receptor (EGFR) has been extensively studied as a potential target for therapy. Overexpression of EGFR is seen associated with up to 60–90 % of high-grade malignant glioma [15], and almost exclusively in de novo high-grade malignant glioma [16]. Grades II, III, and IV astrocytomas have been associated with EGFR positivity with increasing EGFR positivity associated with higher grades of astrocytoma [17, 18]. Several studies have also shown that EGFR is not expressed in normal brain tissue [19, 20]. The 425 anti-EGFR antibody (mAb 425) is an IgG2a isotype developed from mice immunized with A-431 epidermoid carcinoma cells. Compared to contralateral normal brain tissue, in vivo studies showed 10–15-fold localization of mAb 425 to high-grade glioma when nude rats were stereotactically

implanted with human glioma cells [21]. PET-CT imaging with ¹¹¹Indium-labeled mAb 425 of patients with primary gliomas showed 90 % sensitivity, 60 % specificity, and 90 % accuracy to the tumor [22]. After mAb 425 binds to the tumor, anticarcinogenic effects are mediated by direct cell growth inhibition, complement-dependent cytotoxicity, and activation of the humoral response [23–27]. By labeling mAb 425 with radioactive ¹²⁵I, there is a potential for synergistic tumor killing through radiation-mediated DNA damage [25–28]. Survival benefits of adjuvant ¹²⁵I-425 mAb in patients with newly diagnosed high-grade gliomas have been seen in initial clinical studies [29–33]. These prior initial clinical studies included both newly diagnosed GBM and AA and were reported together. The updated results for the GBM have been reported separately [34]. We now report the updated results for the phase II study in which efficacy and safety of adjuvant treatment with ¹²⁵I-425 mAb in patients newly diagnosed with AA were assessed.

Methods

Study design

This present study is an open-label, single-institution, phase II prospective study (NCT00589706) initiated in 1987 to clinically assess the efficacy of adjuvant radioimmunotherapy with ¹²⁵I-425 mAb in patients diagnosed with high-grade gliomas following surgery and external radiotherapy. The study included both newly diagnosed GBM and AA, but our original intent was to study them separately. In this paper, we report the outcomes of patients diagnosed with anaplastic astrocytoma as the GBM results have been already reported previously [33]. The primary endpoint was survival and the secondary endpoint was toxicity. Data regarding progression and progression-free survival were not part of the initial study design and are not available. Decision was made to continue enrollment until phase III trial is available as to allow for continued access to this treatment due to interim analysis showing efficacy.

Patient eligibility

Eligible patients had new diagnosis of histologically confirmed anaplastic astrocytoma (World Health Organization grade III astrocytoma). Exclusion criteria for this study were age <18, Karnofsky Performance Status (KPS) <40, infratentorial tumors, metastatic disease, or second primary cancer. Pregnant or lactating patients and those positive for human anti-mouse antibodies (HAMA) or iodine allergy were also ineligible.

Informed consent was obtained from all patients receiving ¹²⁵I-425 mAb. The study was reviewed and approved annually by the hospital ethics committee and the institutional review board.

Radioimmunotherapy treatment

The 425 monoclonal antibody, provided by Wistar Institute of Anatomy and Biology (Philadelphia, PA), was labeled with 50 mCi (1.8 MBq) of ^{125}I by the Iodogen method as previously described [21, 35] to produce ^{125}I -425 mAb. Administration of ^{125}I -425 mAb were given as three weekly intravenous injections of 50 mCi (1.8 MBq), for a total of 150 mCi (5.4 MBq). Prior to treatments, HAMA assay and delayed hypersensitivity skin testing were done to detect any immunologic reaction to the antibody, and intravenous 25 mg of diphenhydramine was given prior to each radioimmunotherapy to prevent undetected allergic reactions. Patients also received thyroid blockage with daily potassium iodine solution beginning 3 days prior to and until 3 days after treatment with ^{125}I -425 mAb. Surgery, external radiotherapy, and chemotherapy were administered at the discretion of the patient's neurosurgeon, radiation oncologist, and medical oncologist, respectively.

Outcomes and toxicity assessments

Patients have been monitored continually for toxicity and response assessments. For the first 2 h after ^{125}I -425 mAb injections, patients are monitored closely. Initial follow-ups were at monthly intervals up to 6 months, then every 3 months for up to 18 months, then every 6 months. Brain imaging was obtained every 2 months up to 6 months, then every 3 months up to 24 months. The Common Toxicity Criteria for Adverse Events (Cancer Therapy Evaluation Program, National Cancer Institute) [36] was used to assess toxicity.

Statistical analysis

The primary endpoint was survival and secondary endpoint was toxicity. This was an intention to treat analysis, and Kaplan and Meier was used to estimate survival distribution. The power calculation was unable to be done at the time of the design of the study due to lack in literature on the new medication, thus sample size was set arbitrarily at 50 patients. Commercially available program SPSS (version 15) was used for the statistical analysis.

Results

Patient and treatment characteristics

Between 1988 and 2007, radioimmunotherapy with ^{125}I -425 mAb was administered to 80 AA patients at Hahnemann University Hospital as part of this prospective study. The median age was 43 years (range 22–75), the median KPS score was 80 (range 40–100), 46 patients underwent debulking surgery (57.5 %), and 79 patients received radiation therapy

(99 %) with a median dose of 60 Gy. Among the 40 patients (50 %) treated with systemic chemotherapy, 20 patients (25 %) received temozolomide and another 20 patients (25 %) received nitrosourea-based chemotherapy. No chemotherapy was given to 40 patients (50 %). All 80 patients received ^{125}I -425 mAb with a median total dose of 150 mCi (range 42–594) and 11 % of the patients received less than a total dose of 120 mCi. The median time from adjuvant external beam radiation to ^{125}I -425 mAb treatment was 6 weeks. A breakdown of the patient and treatment characteristics is shown in Table 1.

Survival

Sixty-one (76.3 %) of the 80 patients were dead at the time of the analysis; 17 were still alive and 2 were lost to follow-up at the time of analysis. The median overall survival was 55.6 months and the 2- and 5-year survival were 74 and 47 %, respectively (Table 2). The Kaplan–Meier survival curve is shown in Fig. 1.

Toxicity

Infusion of ^{125}I -425 MAb was well tolerated. There was no grade 3 or higher toxicity due to the administration of ^{125}I -425 mAb. Of the 80 patients who were treated with ^{125}I -425 mAb, there were five who experienced acute toxicities.

Table 1 Patient and treatment characteristics

	Treatment group (n=80)
Age	
Median	43
Range	22–75
Sex	
Male	52 (65 %)
Female	28 (35 %)
KPS	
Median	80
≥70	63 (78.8 %)
<70	17 (21.2 %)
Surgery	
Biopsy only	34 (42.5 %)
Debulking	46 (57.5 %)
XRT	
Any	79 (99 %)
Median dose	60 Gy
Chemotherapy	
Nitrosourea-based	20 (25 %)
Temozolomide	20 (25 %)
^{125}I -425 MAb	
Median dose	150 mCi
Range	42–594 mCi

Table 2 Overall survival

	Median survival	95 % CI	Survival at 2 years	Survival at 5 years
RIT	55.6 months	45.9–71.6	74 %	47 %

Grade 1 skin irritation was seen in three patients, grade 1 nausea was seen in two patients, grade 1 infusion-related reaction of transient flushing was seen in three patients, and grade 1 hypotension was seen in one patient. One patient became HAMA positive and received no further treatment. In patients who also received nitrosourea-based chemotherapy prior to radioimmunotherapy or temozolomide during the administration of radioimmunotherapy, no additional toxicity was observed.

Discussion

Radiation therapy remains to be the single most effective postsurgical treatment in patients with AA [37, 38], but despite current standard treatment for AA, recurrences remain high. Accelerated and hyperfractionation schedules, radiosensitizers, systemic chemotherapy, and/or molecular targeted therapy have been pursued in attempt to improve efficacy. Hyperfractionation, accelerated schedules, nor radiosensitizers have shown survival improvements in randomized trials [39, 40].

Chemotherapy use in AA is controversial. The earliest trials to suggest a benefit of chemotherapy in malignant glioma treatment was the early Brain Tumor Study Group trials which showed a trend towards improved survival in the administration of carmustine (BCNU) with radiation [37]. Majority of the previous trials did not separate GBM and AA patients. Adjuvant lomustine, procarbazine, and vincristine (PCV) shortly replaced BCNU after a phase III trial comparing the two regimen showed a survival benefit in those patients who received PCV [7]. However, retrospective analysis of AA patients from four different Radiation Therapy Oncology Group (RTOG) trials showed no survival difference between PCV and BCNU [41]. Furthermore, the Medical Research Council (MRC) phase III trial showed that the addition of PCV had no survival benefit compared to radiation alone [42].

Our phase II study represents the largest reported series using radioimmunotherapy for the treatment of AA. The main goal of this study was to assess the efficacy and toxicity of ^{125}I -425 mAb in patients with newly diagnosed AA. Treatment with ^{125}I -425 mAb was well tolerated and there were no grade 3 or higher toxicities observed. The median overall survival was 55.6 months in those who received ^{125}I -425 mAb in addition to surgery and radiation therapy. Moreover, the combination of ^{125}I -425 mAb and temozolomide or nitrosourea-based chemotherapy was also safe and well tolerated.

Fig. 1 Kaplan–Meier survival curve

Current ongoing study by the RTOG is assessing the effectiveness of combining temozolomide (TMZ) versus combining topotecan, etoposide, and BCNU with radiation in AA patients in the adjuvant setting [12]. However, concurrent chemoradiation carries concerns for higher neurotoxicities. Meta-analysis of AA patients from several prospective studies including RTOG 74-01/ECOG 13-74, RTOG 76-11, and RTOG 79-18 showed that even after adjusting for prognostic factors there was no survival difference between radiation therapy (RT) alone and RT plus chemotherapy [43]. Furthermore, the meta-analysis showed that survival was significantly worse when AA patients also received misonidazole compared to RT alone. Unlike GBM, patients with AA potentially live long enough to manifest the consequences of the treatment despite the poor prognosis. Aggressive treatment regimens, in general, have the potential to be associated with worse survival especially when treatment modalities with higher toxicity profiles are utilized. Extrapolation from GBM studies should be cautioned for this reason. Moreover, extrapolation from AOD and/or AOA is also cautioned given the better prognosis and higher chemosensitivity [5, 7, 41].

This study is subject to the usual biases of an open-label single-arm phase II study and is prone to selection bias. However, when compared to patient composition of other major studies of AA, although median age is similar, patients in this current study had slightly lower median KPS and were less likely to receive optimal surgery. All patients with newly diagnosed AA were offered adjuvant radioimmunotherapy with ^{125}I -425 mAb.

Review of more recently published phase II and III trials of newly diagnosed AA also showed the current study results to be promising in comparison. A PubMed search of phase II and III trials of newly diagnosed AA with more than 30 patients since the year 2000 found five trials with median survival ranging from 14.8 to 63.9 months [6, 8, 9, 42, 44]. Although direct comparison between different studies cannot be made, the current phase II study had comparable promising median survival of 58.7 months in those treated with ^{125}I -425 mAb when compared to these trials. MRC study was excluded since the reported median survival was the worst at 13 months; the remaining four trials along with the current trial are summarized in Table 3.

Accurately classifying malignant astrocytomas histologically can be difficult, but its importance is illustrated by comparing RTOG 94-04 [9] to EORTC 26882 [8]. A central pathology review prior to entry of RTOG 94-04 was done in order to exclude non-AA patients, while in EORTC 26882, the central pathology review was only done after the study had completed. In EORTC 26882, only one third of the cases were confirmed to be AA after review; however, the authors felt this was more of a “real life” scenario. This at least in part explains the lower median survival compared to RTOG 94-04. For our current study, all pathology was reviewed at our academic institution by a neuropathologist, but no formal central pathology review was conducted. Several large AA trials, including EORTC 26882, have found that after central review of the pathology there was greater than 20 % upgrading to GBM [8, 9]. If pathological diagnosis is solely reliant on a stereotactic biopsy then diagnosis would be subject to sampling and the tumor fragment obtained may not represent the entire tumor and would make it more difficult for the pathologist to interpret the tissue. The highest median survival is in the study of Levin et al. [6]. However, the randomization in this study was done after surgery and radiation was completed; likely selecting for those who have completed the standard dose of RT and those who have no evidence of progression. Although the vast majority of patients from this current study received RT, nine patients (11 %) received less than the standard RT dose of 59–60 Gy. It is well known that RT dose can affect survival [45, 46]. Although the current study had the least stringent selection criteria and patients with lower median KPS, it is among the trials with the highest median survivals and lowest toxicity profile.

This study remains one of the most encouraging phase II experiences examining an experimental treatment for AA. Due to its relative low incidence, phase III studies are difficult to do in AA. The median survival of 58.7 months was seen in AA patients who received ^{125}I -425 mAb alone. These results are promising in the setting of poor prognosis and dismal survival and warrants further study.

More recently, several studies have found correlations between genetic aberrations and prognosis in astrocytoma. Those that appear to be relevant in prognosis for AA are tumor suppressor genes involved in *rb1* pathway, PTEN,

Table 3 Direct comparison between different studies along with the current trial

	Study treatment	# of AAF	Median age (years)	Median KPS	Median survival (months)
Current study	^{125}I -425 mAb	77	43	80	58.7
Ron et al. 2002 [44]	PCV	31	50	≥ 70	22
Levin et al. 2003 [39]	PCV +/- DFMO	95 and 86	41.8 and 42.6	(mean) 90.6 and 89.3	63.9 and 41.1 (NS)
RTOG 94-04 [9]	PCV +/- BUdR	134 and 134	<50	90–100	49.2 and 54 (NS)
EORTC 26882 [8]	+/- DBD/BCNU	99 and 94	40 and 44	90	23.9 and 27.3 (NS)

PI3K/Atk/p70s6k proteins, survivin, FPR, MCM3, and chromosome 7. Although these studies are still under investigation, Smith et al. [17] found that AA patients with PTEN gene alterations had significantly worse survival than those without. This association was consistent even after controlling for other clinical relevant factors. Molecular marker analysis was not performed during this study. We are currently analyzing molecular profiles of these patients and the results will be reported separately. Molecular profiles may aid in dividing glioma patients into more homogenous groups, and correlations using ^{125}I -425 mAb treatment outcomes may optimize future treatment regimens.

Conclusion

Treatment with ^{125}I -425 mAb in newly diagnosed AA with or without TMZ or nitrosourea-based chemotherapy is safe and well tolerated. Overall survival is encouraging compared to recently published studies. Further investigations analyzing molecular marker profiles in newly diagnosed AA treated with ^{125}I -425 mAb are currently being pursued.

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Conflict of interest The authors report no conflict of interest concerning the materials or methods used in this study or the findings specified in this clinical article.

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