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The effect of chloroquine treatment in malignant astrocytomas on prognosis

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Aim: To investigate the adjuvant role of the antimutagenic agent chloroquine in the treatment of patients with malignant astrocytomas (MAs).

Materials and methods: Clinical research was conducted at the Neurosurgery Clinic of the Erciyes University School of Medicine, from September 2003 to April 2007, on 37 patients diagnosed with MA after tumor resection subsequent to craniotomy. Chloroquine treatment was started on a daily dose of 150 mg, additional to the patients' radiotherapy and chemotherapy protocol, and was continued throughout the 43-month surveillance period. A control group was formed of 81 patients with MA after the craniotomy.

Results: While the mean survival time of the patients who were treated with chloroquine was found to be 15 months, during the observation period 7 of these patients (18.9%) were alive throughout. In the control group, 20 patients (35.1%) were alive throughout the observation period and their mean survival time was 17 months. There were no statistical differences between the control and chloroquine groups ($P > 0.05$).

Conclusion: The chloroquine treatment was not been found to be effective for the medical treatment of MAs.

Key words: Chloroquine treatment, malignant astrocytomas, prognosis, antimutagenic effect

1. Introduction

In spite of improvements in anaplastic astrocytomas (AA) and glioblastoma multiforme (GBM) diagnostic methods, surgical techniques, and adjuvant treatment, the prognosis of patients with malignant astrocytomas (MAs) is unsatisfactory and is still being expressed in months (1,2). Despite the use of radiotherapy (RT) and /or chemotherapy (CT) as an adjuvant to surgical treatment with these patients, eventual relapse is inevitable. It is known that in these relapsing tumors, mutant clones resistant to RT and CT can develop.

In this study, the adjuvant role of the antimutagenic agent chloroquine, which has an optimum pharmacological profile, in the treatment of MA patients was investigated. The assumption that chloroquine, with its antimutagenic features, added to standard treatment could prevent the development of resistant mutant clones is the basis of this study (3).

In the treatment of various parasitic and immune-based diseases, chloroquine has been used for the last 20 to 30 years. The mechanism of action in GBM treatment is related to either the increase in cytotoxicity induced by conventional treatment or the preservation of mutagenity in neoplastic cells. In this study, the length and dose of chloroquine treatment was planned based on clinical experience obtained from the long-term treatment of autoimmune diseases. The 150 mg/day dose used in studies is accepted as a low dose (4). In addition, it is reported that the dose can be increased up to 300 mg/day and still be well tolerated (5).

2. Materials and methods

Clinical research was conducted at the Neurosurgery Clinic of the Erciyes University School of Medicine, from September 2003 to April 2007, on 37 patients diagnosed with MA after tumor resection subsequent to craniotomy. With demographic, neurological, radiological, surgical,

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and clinical features described prospectively and assumed to affect prognosis, the effect of the addition to adjuvant therapies of 150 mg/day oral chloroquine, an antimutagenic agent, on the therapy and prognosis was investigated in this clinical study.

Informed written consent was obtained from all the patients in the treatment group, and in some cases from their legal custodians. A total of 81 patients, as the control group, diagnosed with MA based on tumor resection after craniotomy and having the same treatment and follow-up criteria except for the chloroquine treatment, were evaluated between November 1999 and August 2003.

2.1. Histopathologic diagnosis

Material obtained through surgical resection was examined by an experienced neuropathologist and categorized as astrocytomas of grades 3 and 4 (AA, GBM) according to the classification of the World Health Organization (WHO) (6). For this group, specially prepared tumor follow-up forms for recording the radiological and clinical features of the patients and treatment methods were completed and kept. The patients' ages, sex, and Karnofsky performance score (KPSs) were recorded throughout the study. Following the examination of their hospital records, all the data related to CT and RT were recorded in their respective forms. While asymptomatic patients were called every 3 months, clinical and radiological data regarding symptomatic cases were recorded on special tumor follow-up forms. At each application for control, a magnetic resonance (MR) test was evaluated by an experienced radiologist who was evaluating the patients for possible relapse. Medicine toxicity was periodically followed up through hematologic, renal, and hepatic function tests. The families of those patients who failed to attend the control sessions were contacted by telephone to determine the neurologic and performance levels of the patients and to invite them to periodic examinations. The death dates of patients who died were obtained from their relatives. The basic criterion for results was the patients' life expectancy.

2.2. Imaging protocol

During the preoperative and postoperative period, enhanced and unenhanced CT and MR imaging tests of all patients were routinely performed. Tumor-related features that were thought to be prognostic were identified by an experienced radiologist. These features were tumor density, localization, diameter, contrasting, extension to opposite hemisphere, clarity of borders, and midline shift. The distance of each tumor to the vital areas in the brain was digitized using the grading system proposed by Sawaya (7) (Table 1).

2.3 Surgical technique

Under general anesthesia, and after the Mayfield Headrest and Skull Clamp System was applied, the position of the patient was fixed using neuronavigation. To refine the surgical deviation resulting from the brain shift throughout the operation, a fusion of intraoperative ultrasound images, with the navigation images adjusted by means of the navigation system, was utilized, and this process was repeated in real-time as required until the end of resection. After resection, intraoperative ultrasound records were obtained for residual tumors when necessary, and when the resection was sufficient, hemostasis was started.

To diagnose any clinically overlooked surgical complication, all the patients underwent CT in the first 4 h after the operation. To compare the degree of tumor resection, and to determine the resection rate on the basis of pre- and postoperative MR examinations, the areas where the signal density increased in a high-contrasted T1 MR examination were accepted as tumor sites. These were identified digitally through the $4/3 \times r1 \times r2 \times r3$ formula and compared with preoperative enhanced T1 A MR images. In the first postoperative 72 h, all the patients were examined by MR and underwent gross total resection. MR findings disagreed with the surgical findings.

2.4. Radiotherapy protocol

While conventional radiotherapy was planned for MA, the surgeon's opinion of the postoperative tumor volume, surgical resection, and pre- and postoperative enhanced CT and MR findings was considered. An effective irradiation area was drawn on the medical mask on the simulator. In the first part of the RT, with a LINAC teletherapy device (Varian 2300 C, USA) at 6,000,000 eV, the area consistent with the tumor, the edematic area around it, and the 2–3 cm brain tissue area outside were identified as the target for the application of radiotherapy, and 40–50 Gy radiotherapy was applied.

Table 1. Grading of malign astrocytomas according to functional location (7).

Grade	Location
I: Nonvital area	Frontal or temporal pole, parietooccipital lobe, cerebellar hemisphere.
II: Near vital area	Near; motor or sensory cortex, calcarine fissure, speech center, internal capsule, dentate nucleus, and brainstem.
III: Vital area	Motor or sensory cortex, visual and speech center, capsula interna, basal ganglia, thalamus, hypotalamus, dentate nucleus, and brainstem.

In the second part of the treatment, the target area was narrowed a little, and the dose was increased to 60 Gy and restricted to 60 Gy in 30 fractions over 6 weeks, the level tolerated by the central nervous system,

2.5. Chemotherapy protocol

After pathological diagnosis, 60 Gy in 30 fractions over 6 weeks was administered. With the first dose of RT, 37 patients were given 75 mg/m^2 of daily oral temozolomide, and the same dose was continued for the 6-week course of RT. After RT, 200 mg/m^2 of 1–5 days of oral temozolomide was given at 4-week intervals, and 6 cycles were completed. The performance scores and hematological, renal, and hepatic functions of the patients were followed closely to make sure they were at normal levels. In cases where aberrant values were found, the CT protocol was interrupted until the values reached normal levels.

2.6. Statistical method

After the operation date, the cumulative lifetimes of the patients were calculated through the Kaplan–Meier method. The survival curve of various inferior groups was compared through the log-rank test. The effect of the multivariables related to patients' survival was analyzed through the Cox regression method. In this study, raw and processed data were calculated within a confidence interval (CI) of 95%. Statistical analyses were performed with the SPSS 10.0 (SPSS Inc., Chicago, IL, USA). Statistical significance was defined at $P < 0.05$ and $P < 0.01$. A statistically insignificant value was defined as $P > 0.05$.

3. Results

3.1. Demographic features of the patients

Of the 37 patients in the chloroquine group, 22 were males and 15 were females with a mean age of 50 years. In 3 of the patients (8.1%), bleeding was detected through tomography as a postoperative complication. In the chloroquine group, relapse developed in 25 patients (80.6%), and 9 of these patients were reoperated on. In the control group, 53 out of 81 patients were male (65.4%), 28 were female (34.6%), and 18 patients were reoperated on. The general features, mean survival time (MST), and statistical differences of the chloroquine and control groups can be seen in Table 2. In the Figure, Kaplan–Meier life curves are shown for the control and chloroquine groups.

3.2. Age and prognosis

The patients were divided into 3 age groups: those below the age of 45 years, those 45–59 years old, and those 60 years old and above. In univariate analyses for the control group, the mean survival time for patients in the group below the age of 45 years was 19 months (CI 95%; range: 12–22), in the 45–59 years age group it was 12 months (CI 95%; range: 9–16), and in the group of age 60 and over it was 6 months (CI 95%; range: 3–9). These findings in

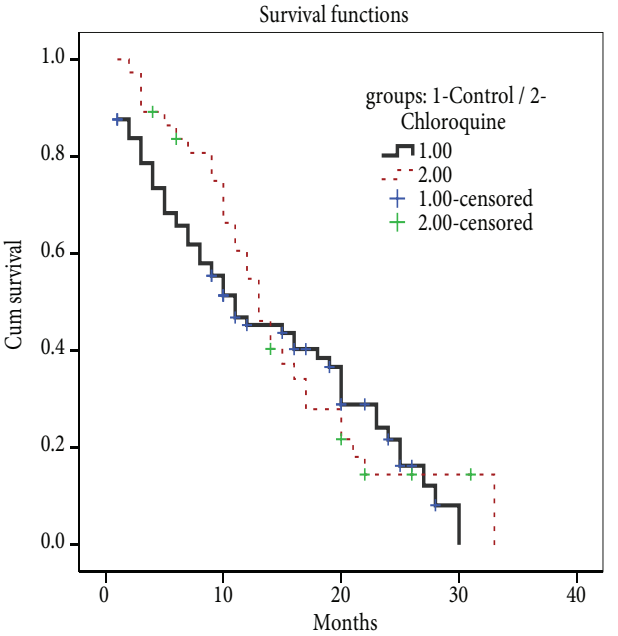

Figure. Kaplan–Meier life curves of the control and chloroquine groups.

the patient group below 45 years old as compared to those in the group of patients age 60 and over were statistically significant ($P < 0.01$), and the younger patients had longer survival times. Compared with the patient group of 60 years and above, the age group of 45–59 years had statistically more significant and longer survival times than those in the group of 60 years and above ($P < 0.05$). In univariate analyses for the chloroquine treatment group, the mean survival time for patients in the group below the age of 45 years was 18 months (CI 95%; range: 13–23), in the age group of 45–59 years it was 15 months (CI 95%; range: 12–19), and in the age group of 60 years and older it was 7 months (CI 95%; range: 4–11). These findings in the patient group of below 45 years and those in the age group of 60 years and older were statistically significant ($P < 0.05$). Patients 45 years old and younger had longer survival times compared with those 60 or older. The age group of 45–59 years had statistically more significant and longer survival times than those in the group of patients of 60 years and older ($P < 0.05$).

The MST of the patients according to their age groups is presented in Table 2. While the younger age groups had statistically significantly longer life expectancy, the differences in survival times and rates in the chloroquine and control groups were found to be statistically insignificant in terms of the prognosis of the disease ($P > 0.05$; Table 2).

3.3. Localization of the tumor and relapse time

In the chloroquine group, 20 tumors were localized in the vital area, 10 in the nonvital area, and in 7 cases near the

Table 2. Demographic and characteristics of patients in the chloroquine and control groups.

Variable		Chloroquine group		Control group		P (between the groups)
		n = 37	Mean survival time (CI 95%)	n = 81	Mean survival time (CI 95%)	
Mean age (range), years		50 (27–72)		50 (21–76)		
Age group	<45	16	18 (13–23)*	33	19 (15–22)ŘŘ	P > 0.05 Ω
	45–59	11	15 (12–19)*	26	12 (9–16)Ř	P > 0.05 Ω
	≥60	10	7 (4–11)	22	6 (3–9)	P > 0.05 Ω
Sex	Male (%)	22 (59.5)	15 (11–19)	53 (65.4)	13 (10–15)	P > 0.05 Ω
	Female (%)	15 (40.5)	13 (9–16)	28 (34.6)	14 (10–19)	P > 0.05 Ω
Localization	Nonvital area	10	16 (9–22)	11 (13.6)	17 (9–25)	P > 0.05 Ω
	Near vital	7	15 (8–22)	22 (27.2)	14 (10–19)	P > 0.05 Ω
	Vital area	20	13 (10–16)	48 (59.3)	12 (9–14)	P > 0.05 Ω
Preoperative Karnofsky score	<70	2	11 (3–18)	27	9 (5–12)	P > 0.05 Ω
	≥70	35	15 (12–18)	54	16 (13–18)ŘŘ	P > 0.05
Operation with neuronavigation		34 (91.9)		67 (82.7)		
Gross total resection (>95%)		31 (83.8)	16 (13–20)**	42 (51.9)	17 (13–20) ŘŘ	P > 0.05 Ω
Subtotal resection (<95%)		6 (16.2)	6 (2–10)	39 (48.1)	10 (8–13)	P > 0.05 Ω
Postoperative Karnofsky score	<70	2	11 (3–18)	35	7 (4–10)	P > 0.05 Ω
	≥70	35	15 (12–18)	46	18 (5–21)ŘŘ	P > 0.05 Ω
Histopathology	GBM	32 (86.5)	15 (11–19)	66 (81.5)	12 (9–14)	P > 0.05 Ω
	AA	5 (13.5)	13 (9–17)	15 (18.5)	20 (14–26)ŘŘ	P > 0.05 Ω
Radiochemotherapy		37 (100)	15 (11–18)	81	17 (15–20)	P > 0.05 Ω
Reoperation		9	17 (13–21)	18	18 (15–22)	P > 0.05 Ω

*: P < 0.05 in the chloroquine group, <45 age group versus ≥60 age group and 45–59 age group versus ≥60 age group.
**: P < 0.01 in the chloroquine group, gross total resection versus subtotal resection.
Ω: P > 0.05 between the groups, chloroquine subgroups versus control subgroups
ŘŘ: P < 0.01 in the control group, <45 age group versus ≥60 age group pre- and postoperative Karnofsky score of <70 versus ≥70 and gross total resection versus subtotal resection, and AA versus GBM.
Ř: P < 0.05, age group 45-59 versus ≥60 age group in the control group.

vital area. In the control group, 48 tumors were in the vital area, 11 in nonvital area, and in 22 cases near the vital area. In inter- and intragroup comparisons of the chloroquine and control groups, tumor localizations in relationship to the vital areas were found to be statistically insignificant in terms of survival times and rates (P > 0.05; Tables 2 and 3). In the chloroquine group, the mean relapse time of the tumor cases localized in the nonvital area was 11 months. The mean relapse time in the case of tumors localized in the vital area was 14 months, and for those localized near the vital area, it was 12 months. The relationship between tumor localizations and relapse times was found to be statistically insignificant (P > 0.05).

In the control group, the mean relapse times for tumors localized in the nonvital area and in the vital area were 11 and 7 months, respectively, while it was 9 months for those near the vital area. The relationship between tumor localization and relapse time was found to be statistically insignificant in the chloroquine and control groups (P > 0.05; Table 3).

3.4. Preoperative and postoperative KPS scores

In the chloroquine group, the patients with a KPS of >70 had a longer life expectancy compared to those with a KPS of <70, the difference between the groups being statistically insignificant (P > 0.05).

Table 3. Relapse time and related factors in gross total resection groups.

Variable		Chloroquine group			Control group		
		n = 31	Survival rate %	Mean relapse time (CI 95%)	n = 42	Survival rate %	Mean relapse time (CI 95%)
Histopathology	GBM	20	15.0	13 (9–17)	33	36.4	6 (4–7)*
	AA	5	0	9 (5–14)	9	66.7	15 (11–19)*
Localization	I	6	16.7	11 (8–14)	7	57.4	11 (5–17)*
	II	9	11.1	12(8–16)	17	41.2	9 (5–13)*
	III	10	10.0	14 (8–20)	18	38.9	7 (4–11)*
Radiochemotherapy		31	18.9	12 (9–16)	42	54.8	11 (9–14)*
Gross total resection (relapse)		25	12.0	12 (9–16)	28	53.6	9 (6–11)*

*: P > 0.05 between the subgroups related with effect on relapse time. Chloroquine subgroups versus control subgroups.
In the chloroquine group, 31 patients had gross total resection rate. In the control group, 42 patients had gross total resection rate. Relapse time and related factors were analyzed in each group. It was detected that there were no statistically significant differences between the subgroups that affected relapse time.

In the control group, the difference between the patients with KPS > 70 and KPS < 70 was statistically significant (P < 0.01) in terms of MST. When the preoperative KPSs of the patients in the chloroquine and control groups were compared in terms of the disease, differences in survival times and rates were found to be statistically insignificant (P > 0.05). There was no statistically significant difference of MST and median survival times between the patient groups with KPS > 70 and KPS < 70 in the chloroquine group (P > 0.05). When the postoperative KPSs of the patients in the chloroquine and control groups were compared, statistically significant differences of survival time and MST were detected in the control group (P < 0.01). When the chloroquine and control groups were compared in terms of the prognosis of the disease, differences in their KPSs and survival times and rates were found to be statistically insignificant (P > 0.05; Table 2).

3.5. Resection level

In the chloroquine group, 34 patients were operated on with the accompaniment of neuronavigation, and 31 patients were determined, based on the postoperative MRI, to have gross total resection. In the control group, 67 patients were operated on with neuronavigation, and their early postoperative MRI recordings revealed that 42 patients (51.9%) underwent total resection and 39 (38.1%) patients underwent subtotal resection. In the control group, relapse developed in 28 (66.7%) patients, and 18 (22.2%) of those were reoperated on (Table 2). In the chloroquine group, when the relapse times of the total resection group and regrowth of the subtotal resection group were analyzed, the mean relapse time was 12 months in the total resection group. In the subtotal resection group, the mean regrowth time was 3 months. The difference between the groups was statistically significant (P < 0.05). In the control group, while the average relapse time was 9 months in the total resection group, the mean regrowth time was 5 months in the subtotal resection group. There was a statistically significant difference between the gross total and subtotal groups (P < 0.05). In terms of the prognosis of the disease, the rates and levels of the resection and the relapse times in the chloroquine and control groups were statistically insignificant (P > 0.05) (Tables 2 and 3).

3.6. Radiotherapy–chemotherapy and relapse time

In the chloroquine group, 25 of the patients receiving RT and CT were detected to have relapse, and 3 of these patients (12.0%) were alive during the follow-up, their mean relapse time being 12 months. In the control group, the mean relapse time was determined at 11 months in RT- and CT-receiving patients. In the chloroquine and the control groups, in terms of the prognosis of the disease, the relationship between RT and CT and relapse time was found to be statistically insignificant (P > 0.05; Table 2).

3.7. Pathological diagnosis and relapse time

The histopathological diagnoses and the relapse times of the patients treated with chloroquine were examined. In the chloroquine group, the mean relapse time of the patients with AA was determined at 9 months, while it was 13 months for the patients with GBM. The difference between the histopathological subgroups was found to be statistically insignificant (P > 0.05). Table 3 presents Kaplan–Meier survival curve information of the patients in the chloroquine group based on their pathological

diagnosis and the relapse time. In the control group, the mean relapse time of the patients with AA was 15 months, but for those with GBM it was determined at 6 months. The difference between the relapse times of the histopathological subgroups was found to be statistically significant ($P < 0.01$).

The pathological diagnosis and the relapse time rates in the chloroquine and the control groups were found to be statistically insignificant ($P > 0.05$) in terms of the prognosis of the disease (Table 3).

4. Discussion

The prognosis of GBM patients has changed little in the last decade. The most recent studies show that after an aggressive treatment consisting of a combination of surgical therapy, radiotherapy, and chemotherapy, the average life expectancy of the patients is about 1 year (1).

Chloroquine firmly binds to nucleic acids, especially to DNA's cytosine–guanine sequence. It tonifies the structural configuration of DNA and prevents mutagenesis. Chloroquine leads to stabilization of the p53 protein and induces the p53 transcriptional target in glioma cell lines with wtp53. The p53 independent cytotoxic effects of chloroquine are well known and are related to the ability of chloroquine to cause mitochondrial dysfunction as a result of the inhibition of lysosomal autophagy (2).

This effect has been shown in many eukaryotic cells, including viruses, bacteria, and various cancer cells (8,9). Apart from this important feature, chloroquine, a necrosis factor, acts almost like an immunomodulatory by inhibiting phospholipase A2 and tumors (10). Furthermore, it impairs the DNA cell repair mechanism that results from the degradation induced by alkylating treatment (11).

In vitro, chloroquine blocks the antigenic protein expression on the surface of malignant glial cells and penetrates these cells. Because of its reducing effect on the DNA's cell repair mechanism and DNA synthesis, chloroquine strongly reveals the inhibitive effect of radiation on cell proliferation. Although chloroquine alone is a cytotoxic, when accompanied by ionizing radiation, it causes subsequent changes in cell structure such as carcinoma and melanoma, as well as serious ultrastructural lesions characterized by microtubule and microfilament increase, mitochondrial damage, and endoplasmic reticular vacuolization (12).

Furthermore, other intracellular effects of chloroquine can increase the irritability of malignant glioma cells to the standard treatment. Because of the elevation of endozoic and lysosomal pH, which increases and sustains the concentration of the lipophilic antineoplastic drugs like carmustine, it increases the permanence of cancer cells (13).

The remarkable effect of chloroquine, which somehow effectively reverses the multidrug resistance of cancer cells, is that it delays or prevents the extracellular transportation of anticancer medications like vincristine (13). Finally, the addition of chloroquine to leukemic cell cultures with multidrug resistance has been able to reduce the resistance to vinblastine by between 10- and 15-fold (13,14).

Malignant cell clones that are resistant to CT and RT cause treatment failure in GBM patients. It is because of this resistance that the addition of new treatments to the conventional treatment is thought to improve results (15).

High mutagenesis in malignant cells is a primary factor in determining whether cell clones are resistant to chemotherapy. Quinacrine binds firmly to DNA, preventing mutagenesis. In a study on rats, Reyes et al. (16) reported that quinacrine added to carmustine treatment increased the antineoplastic effect of carmustine and prolonged its effectiveness. According to their conclusions, the inhibition of mutagenesis in malignant glial cells during chemotherapy prevents the emergence of resistant clones.

Because of all these effects, chloroquine has been thought to contribute to recovery in GBM treatment.

Briceno et al. (17,18) reported that chloroquine, which has an antimutagenic profile, considerably increases MA response to antineoplastic therapy. This finding has been ascribed to the antimutagenic effect, which prevents the emergence of resistant clones during RT and CT.

Briceno et al. (17,18) also noted that chloroquine treatment affects the patients' MST and relapse time positively. In our study, however, the difference between the chloroquine and control groups was not statistically significant ($P > 0.05$). In inter- and intragroup comparisons, the pathological diagnosis and relapse time rates were found to be statistically insignificant in terms of the prognosis of the disease ($P > 0.05$). Our findings do not support those of Briceno et al. (17,18).

There is little information about the effects of the age factor in MA on radiosensitivity. Kumabe et al. (19) found that 22 patients with malignant glioma and complete reaction to RT were younger than those with less reaction. In another study (20), the mean age of 11 GBM patients who showed complete reaction to the radiation was 42. Even this age is younger than that of GBM patients in most studies (21).

Barker et al. (22) detected that, along with variables such as extended surgical resection and high preoperative KPS, the younger age of the patient also enhances the response to radiation. There is important evidence that shows a relationship between the age and longer life expectancy in adult patients with MA. Mortality risk increases with age. Aggressive treatment is less effective in older patients than in younger patients in terms of

prolonging life expectancy (22). In the present study, statistically significant differences of survival were found in the chloroquine group between the patients aged below 45 years and the age group of 45–59 years ($P < 0.05$). In the control group, however, a statistically significant longer life expectancy was detected in the patients aged below 45 and in those in the age group of 45–59 years ($P < 0.01$ and $P < 0.05$, respectively). In terms of the prognosis of the disease, an intergroup comparison of the survival times and rates did not show any statistical difference ($P > 0.05$). These findings in the control and study groups are consistent with the literature data.

The 2 clinical variables that determine the radiation reaction in MA patients are the KPSs of the patients before RT and the degree of surgical resection (22). Curran et al. (23) argued that MA patients with superior performance and more comprehensive resections responded to RT more favorably. It has been reported that high tumor metabolism can be detected through positron emission tomography screening of the MA patients with low KPSs (24).

In some studies on the effect of surgery on life expectancy, the findings have been in favor of aggressive tumor resection (22,25).

In this study, inter- and intragroup comparisons of the effect on MST of the tumor localization in relationship to the vital areas were also performed. Although MST was detected to be longer in cases of tumors in nonvital areas, this finding was statistically insignificant ($P > 0.05$). However, the findings are of a nature to corroborate the findings from previous studies.

The effect of surgery on immunotherapy can emerge through the reduction of tumor load and a mechanical disorder in the blood–brain barrier (26). In numerical studies on tumor size in patients with recurrent MA, it has been noted that when the postoperative size of the tumor is small, survival is significantly longer (27). Theoretical tumor models indicate that smaller-sized tumors can show greater radiosensitivity (28).

Salcman (29) defended the validation of comprehensive surgery based on the literature screening of selected patients who did not have adjuvant therapy, even if he did not allow for tumor localization in his analyses.

In the literature (22,25), along with the studies that indicate that the bigger the resection is, the longer the lifetime will be, there are also some studies that show that the degree of resection has no effect on survival rates (30). On the other hand, Iliadis et al. (30) reported that net-enhancing tumor volume before radiochemotherapy was a statistically significant independent variable in the multivariate Cox analysis. However, in the studies on general prognostic factors, the extent of resection is the more often observed factor that has a desirable effect on survival time (22,25).

In this study, a statistically meaningful difference between the chloroquine group and the control group was not determined when considering MST ($P > 0.05$). However, in intragroup comparisons, the total resection patients in both the control and treatment groups had longer lifetimes compared to those in the subtotal resection group. In terms of the prognosis of the disease, the degree of resection and the regrowth time were found to be statistically significant in intragroup comparison ($P < 0.05$), but not in intergroup comparisons ($P > 0.05$). Even though these findings support the literature findings that aggressive surgical resection improves the prognosis, it shows that chloroquine treatment does not have a favorable effect on regrowth.

Curran et al. (23) found that radiation reaction was assessed as better in terms of image in MA patients, with better performance and more comprehensive resections. In this study, survival times and rates were found statistically insignificant in patients with RT in intergroup comparisons ($P > 0.05$). This finding is far from supporting the thesis and main idea of this study, that chloroquine treatment could be effective in maintaining the irritability of tumor cells to RT.

Sotelo et al. (5) reported that survival advantage could be achieved in the medium term in the chloroquine and control groups, but the long-term prognosis is poor.

Our findings do not support the findings of Reyes (16) and Briceno et al. (17,18), and chloroquine treatment has not been found effective in increasing sensitization to RT and CT.

The addition of adjuvant treatments, such as CT and RT, to the surgical treatment of GBM can increase patients' life expectancy in the medium term. However, the long-term prognosis is still far from satisfactory. The data regarding chloroquine, with its antimutagenic feature that has been established in laboratories and also the remarkable improvement it provides in the prognosis of the patients through clinical usage, have been the basis for the planning stage of this study. However, the results we obtained do not support the previous studies on this subject, and they are disappointing in terms of the prognosis.

Finally, contrary to previous studies, chloroquine has not been found to increase the response of GBM to antineoplastic treatment. Our findings do not support the findings of the previous studies, and chloroquine has not been found effective for the medical treatment of high-grade astrocytomas. Further studies on larger patient groups, with dose comparisons, could clarify the subject.

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