High-Dose Methotrexate for Intraocular Lymphoma

Tracy T. Batchelor, Gina Kolak, Roberto Ciordia, C. Stephen Foster, and John W. Henson

Massachusetts General Hospital Brain Tumor Center and Neurology Service, Massachusetts General Hospital, Boston, Massachusetts 02114 [T. T. B., G. K., R. C., J. W. H.]; Department of Neurology, Harvard Medical School, Boston, Massachusetts 02140 [T. T. B., R. C., J. W. H.]; and Department of Ophthalmology, Massachusetts Eye and Ear Infirmary, Boston, Massachusetts 02114 [C. S. F.]

ABSTRACT

Purpose: Intraocular lymphoma (IOL) frequently coexists with primary central nervous system lymphoma (PCNSL). We sought to determine the efficacy of high-dose methotrexate (MTX) alone in patients with IOL. We also sought to determine whether micromolar levels of MTX could be achieved in the aqueous and vitreous humor of the eye after i.v. administration of the drug.

Experimental Design: Nine patients with concurrent PCNSL and IOL or isolated IOL were treated with MTX alone. All patients were treated with i.v. 8 g/m² MTX. MTX concentrations in serum, aqueous humor, and vitreous humor were obtained in seven of nine patients with IOL and in one additional patient with PCNSL but no evidence of IOL.

Results: Micromolar concentrations of MTX were present in both ocular chambers 4 h after completion of the infusion in eight of eight patients. Levels of MTX were lower in the vitreous humor compared with the aqueous humor in five of six patients in whom both chambers were assayed. Initial response of IOL to MTX was demonstrated by seven of nine patients (six complete responses and one partial response), whereas two patients had persistent IOL despite achievement of micromolar concentrations of MTX. In the patients with concurrent PCNSL and IOL, seven of seven had complete responses in the brain after treatment with MTX. Three of seven patients with initial response of IOL experienced relapse in the eye requiring orbital radiation, and four of nine patients had sustained response of IOL to MTX.

Conclusions: A subset of patients with IOL may experience sustained remission when treated with high-dose i.v. MTX alone. Although micromolar MTX concentrations are present in the eye 4 h after infusion, the lower concentration achieved in vitreous humor may contribute to persistence of IOL.

INTRODUCTION

IOL refers to infiltration of the vitreous humor, retina, and choroid by malignant lymphocytes and was first described in 1951 (1). Characteristic signs in the vitreous humor include sheets or clumps of cells on vitreous strands and vitreitis with multifocal subretinal pigment epithelial infiltrates (2). Other signs include multiple deep white dots in the retina secondary to tumor infiltration, necrotizing retinitis, infiltration of the retinal vasculature causing arterial or venous obstruction, and optic nerve invasion (3). The median age at diagnosis of IOL is 60 years, and there is no clear gender predilection (2, 4, 5). Bilateral involvement occurs in up to 90% of patients (4, 5). IOL may precede or occur synchronously with PCNSL. In one report, 16 of 20 (80%) patients with IOL had involvement of the CNS at some point in the course of their illness (5). Extra-CNS involvement was reported in 22% of IOL patients in another case series (4).

The presence of uveitis in a patient with known PCNSL is highly suggestive, but not diagnostic, of IOL. Blurred vision, diminished visual acuity, and floaters are the most common symptoms of IOL, although some cases of IOL are asymptomatic. A high index of suspicion must be maintained for IOL because early visual symptoms are nonspecific. The delay from the onset of symptoms to diagnosis of IOL has been years in some studies (4). Uveitis that is resistant to treatment with steroids should raise the clinical suspicion of IOL (6). Treatment with steroids before acquisition of pathological material may decrease the diagnostic yield of vitreal biopsy (6).

The slit lamp examination is abnormal in more than 90% of IOL cases, revealing uveitis, vitreitis, and retinal or choroidal infiltrates. The identification of malignant lymphocytes in vitreous humor remains the definitive method for diagnosis of IOL, and vitreal biopsy establishes the diagnosis of IOL in 95% of cases (5). Tumor cells are typically large and pleomorphic with round or oval nuclei with nuclear membrane irregularities and one or more micronucleoli (4). Other methods that have been successful in the diagnosis of IOL include the use of vitreous washing specimens for the detection of B-cell and T-cell gene rearrangements by PCR and elevation of the interleukin 10 to interleukin 6 ratio (>1.0). Noninvasive tests such as ophthalmic ultrasonography are under investigation (6–9).

This previously rare ocular disease has become more frequent partly because of the 3-fold increase in incidence of PCNSL since the early 1970s (10). The mechanism by which
malignant lymphocytes communicate between the eye and the CNS is unknown. However, direct invasion of the optic nerve and meningeal infiltration are two possible routes of CNS dissemination (6). The etiology of IOL is unknown. Some studies have detected EBV and human herpes virus-8 DNA and Toxoplasma gondii in vitreous specimens or ocular tissues obtained at the time of autopsy (11).

Ocular irradiation has been the main treatment modality for IOL. However, PCNSL is not prevented or treated with this local form of therapy, and the potential for significant toxicity exists. We therefore analyzed the response of IOL in immunocompetent patients to treatment with i.v. MTX at a dose of 8 g/m² and sought to determine whether cytotoxic intraocular concentrations of MTX could be achieved after i.v. administration of this drug.

PATIENTS AND METHODS

All patients were treated with the same high-dose MTX schedule (12). Chemotherapy consisted of induction (8 g/m² MTX administered every 14 days until complete response), consolidation (8 g/m² MTX administered every 14 days for 2 doses), and maintenance (8 g/m² MTX administered every 28 days for 11 doses) phases. Doses of MTX were adjusted on the basis of reductions in creatinine clearance. A minimum creatinine clearance of 50 ml/min was required before treatment with MTX. Additional requirements included a neutrophil count ≥1,500/mm³ and a platelet count ≥100,000/mm³. All patients were admitted to the hospital for each cycle and received prechemotherapy antiemetics, urine alkalization, and i.v. hydration. Alkalization and i.v. hydration were continued until the patient achieved a urine pH of ≥7 and a urine output of 100 ml/h for 4 consecutive hours. After these parameters were achieved, patients received i.v. ondansetron at doses ranging from 8–24 mg. Thirty min later, the MTX infusion was started and continued for a total of 4 h. During this time, i.v. hydration and alkalization were continued to maintain the parameters outlined previously. Twenty-four h after the start of the MTX infusion, calcium leucovorin was administered p.o. at a dose of 25 mg every 6 h. During each cycle, daily plasma MTX levels were obtained, and patients were discharged when the plasma MTX level was <0.10 μM. Patients were hospitalized for a median of 4 days for each cycle of chemotherapy.

All patients were followed via serial eye examinations by a single ophthalmologist. Vitreous and aqueous samples were obtained for cytopathological analysis in seven of nine patients with IOL and in another patient with PCNSL but no clinical evidence of IOL. Vitreous and aqueous samples were obtained 4 h after completion of the MTX infusion. Simultaneous plasma MTX levels were obtained. In three patients, a CSF sample was also obtained immediately before or after the vitreal biopsy, and MTX was measured in this fluid.

MTX concentrations in serum, vitreous humor, aqueous humor, and CSF were measured using an enzyme-multiplied immunoassay (Syva Company, Cupertino, CA). To check the accuracy of the test in the vitreous humor, sample dilutions with saline were done and analyzed along with undiluted samples in patients 1, 4, and 5. The linear relationship of the vitreous concentrations in the dilutions supported the validity of the assay.

RESULTS

Over the course of the study, seven immunocompetent patients with concurrent CNS lymphoma and IOL (five patients with PCNSL and two patients with CNS metastases) and two patients with isolated IOL were treated with i.v. MTX at a dose of 8 g/m², after giving informed consent. There were five males and four females with a median age of 52 years in this case series (Table 1). In five of nine patients, IOL was diagnosed concurrently with brain or leptomeningeal lymphoma. In three of eight patients, IOL was diagnosed 8 months to 4 years before the development of neurological symptoms and diagnosis of PCNSL. One patient was diagnosed with isolated IOL. Initial diagnosis of IOL was determined by the presence of malignant or atypical lymphocytes in the vitrectomy specimen in seven patients and by ophthalmic examination in two patients who presented with ocular symptoms and positive slit lamp examination in the setting of PCNSL and CNS metastases from systemic non-Hodgkin’s lymphoma. In one of the latter patients, a subsequent vitreal biopsy documented malignant B lymphocytes in the specimen. All patients gave informed consent before vitrectomy. Staging for all patients included computed tomography scans of the chest and abdomen, HIV serology, and a lumbar puncture with CSF flow cytometry and cytopathology. Two patients had atypical cells identified in the CSF. One patient had a history of testicular lymphoma, but this had responded to prior chemotherapy, and there was no evidence of disease elsewhere in his body when he presented with ocular and brain lymphoma. Another patient had prior stage IV non-Hodgkin’s lymphoma and residual adenopathy at the time a cerebellar mass was discovered. Biopsy of the brain lesion revealed diffuse, large B-cell lymphoma.

After treatment with MTX, seven of nine patients had a response in the eye, and two of nine had refractory IOL requiring orbital radiation. Both of the patients treated with orbital radiation for refractory IOL had complete responses to this modality of treatment. Of the seven patients who had a response, four have had sustained responses in the eye at follow-up after 8+, 15+, 20+, and 36+ months, whereas three experienced recurrent IOL 6, 17, and 24 months after initial diagnosis of IOL. All seven patients with concurrent PCNSL and IOL had complete responses of their brain disease. Six patients were alive 8+, 8+, 15+, 20+, 36+, and 85+ months after diagnosis of IOL. One patient died 17 months after diagnosis of IOL, and another died 27 months after diagnosis of IOL, each from CNS progression. One patient was lost to follow-up.

Initial resistance of IOL to MTX or subsequent ocular relapse after initial response of IOL to MTX did not appear to be a poor prognostic marker because three of four patients responded to orbital radiation. One patient with recurrent IOL after orbital radiation responded to i.v. MTX on two subsequent occasions. Toxicity related to MTX was modest, with only two episodes of grade 3 anemia, one episode of grade 3 neutropenia, and no treatment-related deaths. All patients were followed with serial mini-mental status examinations, and the posttreatment scores obtained at last follow-up are shown in Table 1. On the basis of these scores, it can be inferred that neurotoxicity was minimal for all patients despite the presence of brain or ocular lymphoma and treatment with high-dose MTX.
The results of the serum, CSF, vitreous humor, and aqueous humor concentrations from eight patients are shown in Table 2. In all eight cases, the vitreous and/or aqueous concentrations 4 h after MTX infusion were higher than 1 μM, levels usually considered cytotoxic and capable of eradicating lymphoma cells in the CSF. Micromolar concentrations of MTX were achieved in the CSF in all three patients who underwent lumbar puncture at the time of the vitrectomy. There were too few cases for meaningful statistical analysis of predictors of response and survival. There were no statistically significant correlations of serum and ocular (aqueous and vitreous humor) MTX levels.

### DISCUSSION

The prognosis for patients with IOL is generally poor because most develop CNS dissemination (4, 6). Standard treatment of IOL consists of focal radiation to the eyes, and local control is achieved in most cases. Standard doses to the orbit have ranged from 30–45 Gy (3, 5). In one series of eight patients treated with irradiation (ranging from 35–45 Gy), vision improved in 10 of 13 eyes. However, most patients relapsed in the CNS and died 5–39 months after diagnosis (13). Thus, although local control is satisfactory after ocular irradiation in most cases, this approach has three disadvantages. First, PCNSL is not prevented or treated with this local form of therapy. If no additional treatment is provided, approximately 90% of these patients relapse or develop PCNSL (10). Second, the potential for significant toxicity (cataract formation, retinal detachment, and optic nerve atrophy) exists. Third, the treatment usually cannot be repeated if the patient relapses.

Intravitreal MTX is another form of local therapy for IOL. In one series of 16 IOL patients, 26 affected eyes were treated with intravitreal injections of MTX (400 μg) twice weekly. This injection schedule is supported by a study of the ocular pharmacokinetics of intravitreal MTX in rabbits. In this study, it was found that a single 400-μg intravitreal dose of MTX results in therapeutic (>0.5 μM) levels of the drug in the vitreous humor for 48–72 h (14). All eyes were cleared of malignant cells with this regimen. Relapse requiring retreatment with intravitreal MTX occurred in 3 of 16 patients. All deaths in this study were attributable to progression of intracranial lymphoma. Toxicity from this regimen was significant and included cataract formation (69%), corneal epitheliopathy (56%), retinal pigment epithelial disturbance (38%), vitreous hemorrhage (13%), sterile endophthalmitis (6%), and optic atrophy (6%). These complications led to deterioration of visual acuity by 2 or more Snellen lines in 6 of 16 patients (15). Even though intravitreal MTX is associated with a high initial response rate, major disadvantages of this local form of therapy include toxicity and failure to treat possible microscopic disease in the brain or CSF.

The first well-documented report of the successful treatment of IOL with systemic chemotherapy involved a 77-year-old woman with recurrent IOL treated with procarbazine, 1-(2-chloroethyl)-3-cyclohexyl-1-nitrosourea, and vincristine. Ultrasonographic imaging demonstrated resolution of the tumor with this treatment (16). Since that time, several case series using Ara-C have been reported (17–19). In one case, i.v. Ara-C at 3 g/m² resulted in micromolar concentrations in the aqueous and vitreous humor 90 min after the infusion. Several responses of IOL to Ara-C alone or in combination with other drugs have been reported (17–19). However, in one series, a complete response was observed in only one of five cases (18).

High-dose i.v. MTX is a successful treatment for PCNSL, and prior anecdotal reports have shown that micromolar concentrations can be achieved in the vitreous humor after this therapy (20, 21). Conversely, intrathecal MTX does not lead to...
detectable levels of the drug in the eye (20). Although it remains controversial, it has been reported that 1 μM is an effective concentration of MTX in CSF for the treatment of hematological malignancies that have disseminated to the leptomeninges (22). A study of the in vitro cytotoxic activity in 63 different cell lines has demonstrated that therapeutic levels of the drug range from 0.1–1 μM with a mean IC₅₀ of 0.32 μM (14). However, as demonstrated by our case series, micromolar concentrations may not be sufficient to eliminate IOL in all cases. An alternative explanation for the treatment resistance of some of our cases may be insufficient duration of exposure to MTX at these cytotoxic levels. All MTX measurements were made 4 h after the i.v. infusion, and we do not have information on levels at subsequent time points. Initial resistance of IOL to MTX or subsequent ocular relapse after initial response of IOL to MTX does not appear to be a poor prognostic marker because three of four patients responded to orbital radiation. One patient with recurrent IOL after orbital radiation responded to i.v. MTX on two subsequent occasions.

In other studies of high-dose i.v. MTX, anecdotal responses have been reported for newly diagnosed concurrent PCNSL and IOL or IOL alone. In a cooperative group study of newly diagnosed PCNSL, five patients who also had IOL were treated with the same regimen used in this study (8 g/m² MTX every 14 days) with complete responses in the brain in five of five patients and resolution of vitreal cell infiltrates in four of four patients (12). These data suggested that high-dose MTX may be an effective initial treatment for IOL associated with PCNSL. In another study of MTX (8.4 g/m² over 24 h), thiotepa (35 mg/m²), vincristine (1.4 mg/m²), and dexamethasone for newly diagnosed PCNSL, five patients had concurrent IOL. Of these five patients, three had complete responses in the brain and eye after treatment with this regimen (23). On the basis of these preliminary results, it appears that MTX-based chemotherapy may be a viable initial treatment option in patients with IOL and PCNSL with IOL.

In a study of high-dose chemotherapy followed by hematopoietic stem cell rescue in patients with relapsed PCNSL or IOL, 12 of 22 patients had IOL or PCNSL with IOL at the time of diagnosis. After treatment, 9 of 12 patients had complete responses of the brain and eye disease to chemotherapy, and 9 of 12 patients were alive 21–84 months after relapse (24). However, infectious complications were common (19 of 22 patients), and 7 of 22 patients developed neurotoxicity (acute encephalopathy, 2 patients; severe chronic encephalopathy, 5 patients), resulting in death in 2 patients. Moreover, five of seven patients >60 years of age died from treatment complications in this study (24). Given the high proportion of severe toxicities, it is not likely that this treatment strategy will be widely adopted in this patient population.

In summary, micromolar concentrations of MTX are reliably achieved in the vitreous humor, aqueous humor, and CSF after treatment with i.v. MTX (8 g/m²). Despite the establishment of presumably cytotoxic concentrations of MTX in vitreous and aqueous humor, IOL can persist after 2–3 cycles of high-dose MTX and may relapse after initial response to i.v. MTX.

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