vitreous cavity. Strong glial fibrillary acidic protein positivity confirms the diagnosis. Thickening of the vessel walls, perivascular pigment, intralesional cysts, and calcareous deposits (calcospherites) are also detectable.

Massive retinal gliosis is characteristically encountered in phthisical eyes after trauma, surgery, or inflammation and in other conditions such as retinopathy of prematurity or Coats disease. Our patient’s MRG was probably a poorly modulated reparative response to an unexplained preceding hemorrhage in the eye. The smaller retinal vasoproliferative tumors are also seen in such settings, implying that they too are usually reactive. We doubt there is any intrinsic retinal property in NF1 that is conducive to MRG. Studies of MRG have preliminarily shown polyclonality, further supporting a reactive lesion. Immunostains for p53 and Ki-67 here are close to negative, whereas they are positive (>10% of cells) in astrocytic lesions. Immunostains for p53 and Ki-67 here are close to negative, whereas they are positive (>10% of cells) in astrocytic lesions. Other conditions such as retinopathy of prematurity or Coats disease.

Subconunctival Indocyanine Green Identifies Lymphatic Vessels

The lymphatic system maintains tissue fluid balance and immunity. It also plays an important role in pathologic conditions such as tumor metastasis and inflammation. The anatomicists Arnold (in 1847) and Teichmann (in 1861) were the first to visualize and describe the conjunctival lymphatics. In 1948, Busacca identified a number of anastomosing lymphatics, which penetrated the orbital cavity near the extraocular muscles, using trypan blue dye. A better understanding of the lymphatic system in the eye could provide the basis for developing alternative therapeutic strategies for ocular diseases.

Newer techniques of fluorescence lymphography using indocyanine green (ICG) are increasingly being used in medical specialties such as vascular surgery and oncology. Lymphography using ICG is useful because there is no endogenous fluorescence in the near-infrared band (780-1500 nm) used for ICG detection and the examination does not cause tissue damage. Indocyanine green is approved for hepatic and ophthalmologic applications. We identified conjunctival lymphatic capillaries using lymphography with ICG.

Methods

This study was approved by the independent and external Hospital Agamenon Magalhaes Institutional Review Board. One of us (C.A.F.-N.) volunteered as the study participant and provided written informed consent. One vial containing 5 mg of ICG used for internal limiting membrane staining (Ophthalmos) was used to prepare a sterile aqueous ICG solution at 5%. Following topical anesthesia, 0.1 mL of this ICG solution was injected into the subconjunctival area using a 30-gauge needle through the conjunctiva approximately 3 mm away and inferotemporally from the limbus of the right eye.

Ocular imaging was performed using a confocal scanning laser ophthalmoscope (Spectralis HRA + OCT; Heidelberg Engineering Inc). For color photography, a retinal camera (TRC-50IX; Topcon Medical Systems) was used. Ophthalmic evaluation and ocular imaging were performed on a weekly basis until complete absorption of the ICG was verified.

Results

No systemic or local adverse effects were observed following the subconjunctival injection of ICG. The ICG completely cleared from the eye within 4 weeks. Minute lymph vessels (lymphatic capillaries) were identified and differentiated from blood vessels by multimodality diagnostic imaging. Metabolic analysis of the ICG lymphography revealed intermittently dilated lymphatic drainage channels under the conjunctival blood vessels (Figure).

Discussion

Three decades ago, Rayes et al studied the lymphatic distribution in the bulbar conjunctiva of 60 patients by injecting 1% trypan blue dye in different quadrants of the bulbar conjunctiva. The scheme of the lymphatic distribution in normal bulbar conjunctiva was presented and histopathologic studies confirmed the presence of lymphatic tissue. During phacotrabeculectomy surgery, Singh observed several vessels through which the dye passed after subconjunctival injection of trypan blue dye. The conclusion was that there...
were likely lymphatic vessels. These were observed in more than 120 cases of all types of glaucoma. Singh suggested that the excess interstitial subconjunctival fluid is removed by the conjunctival lymphatic system. We present initial data about the use of ICG to identify the microanatomy of the ocular lymphatic system in humans. After a single subconjunctival injection of 0.5 mg of ICG, lymphatic vessels were clearly identified on near-infrared fluorescence. 

The lymphatic drainage of the eye remains undefined. Unexpectedly, we could also document that ICG hyperfluorescence drained toward the perilimbal region. The ICG remained in the corneal avascular tissue for several days before complete disappearance. Further topographic mapping studies with near-infrared fluorescence lymphography are needed to better understand normal human conjunctival lymphatic microanatomy and drainage.

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Intralesional Rituximab for Primary Iris Lymphoma

Primary B-cell mucosa-associated lymphoid tissue (MALT) lymphoma of the iris is rare. The largest published series of 14 iris lymphomas included 3 such lymphomas.1 Thus, data concerning the effectiveness of treatment of these tumors are limited. External radiotherapy (the main treatment), resection, and systemic chemotherapy can cause substantial morbidity. Rituximab monoclonal antibody, targeting CD20 on B cells, has been used systemically and intralesionally to treat orbital and primary intraocular lymphomas and ocular inflammatory diseases.2,3 To our knowledge, we report the first case of primary iris MALT lymphoma treated with intralesional rituximab injection.

Report of a Case | A man in his late 60s presented for evaluation of a right iris lesion that had been noted during routine examination. Visual acuity was 20/25 OU. Slitlamp examination showed a 10 × 9 × 3.5-mm, well-delineated, bilobed, vascularized, tan, peripheral iris nodule between the 3- and 6-o’clock positions (Figure, A). There were episceral sentinel vessels and anterior chamber angle involvement but no visible iris seeding or aqueous cells (Figure, B). The eye was pseudophakic. Ciliary body involvement was seen on ultrasonographic biomicroscopy (Figure, C). Fine-needle aspiration biopsy showed a monomorphic population of small to medium-sized hyperchromatic, CD20+ lymphocytes of MALT lymphoma (Figure, D and E). Few CD3+, reactive T lymphocytes were present (Figure, F). Systemic evaluation showed no other involvement. The tumor was staged as T2N0M0.

Treatment options of external radiotherapy, brachytherapy, and excision were discussed, but the patient elected intralesional rituximab based on successful treatments of primary intraocular and orbital lymphomas with rituximab injections. Using a 32-gauge needle, an anterior chamber tap was performed to release 0.05 mL of aqueous humor. Using another 32-gauge needle, 0.1 mL of rituximab (1 mg/0.1 mL) was injected into the tumor through the cornea infratemporally under slitlamp visualization. In total, 3 injections, repeated monthly, were performed. Beginning with the first injection, the tumor shrank and completely disappeared on slitlamp, gonioscopic, and ultrasonographic biomicroscopic examinations by 5 months (Figure, G-I). There were no anterior segment toxic effects or recurrence 8 months after the last injection and no evidence of systemic lymphoma based on examination and fludeoxyglucose F 18 positron emission tomography.

Discussion | Intralesional rituximab injection is used successfully to treat ocular lymphomas. Savino et al4 described 7 patients with adnexal MALT lymphomas, treated with 4 weekly injections and followed up for more than 1 year, who had complete remission (4 patients [57%]), partial response (2 patients [29%]), or no response (1 patient [14%]); the disease remained stable for 4 years in the patients with partial responses. Ferreri et al5 reported complete responses to rituximab injections for 2 of 3 conjunctival MALT lymphomas, without adverse effects.

We found intralesional rituximab to be an effective treatment for primary iris lymphoma, but long-term effectiveness is unknown. Reduced effectiveness might occur for high-grade lymphomas or those with ciliary body and choroidal involvement. Potential complications of intralesional injection are hyphema, cataract, increased intraocular pressure, dispersion of tumor cells, and corneal toxic effects. Our case was diagnosed using fine-needle aspiration biopsy, which has not been shown to increase the risk of seeding or metastasis.1

We used the same dose and frequency of rituximab as used for primary intraocular lymphoma. Pharmacokinetics in rabbits show that rituximab’s half-life is 4.7 days in the vitreous and 5.3 days in the aqueous humor after an intravitreal injection, indicating that at least 10 μg remains in the eye 30 days after a 1-mg injection,6 with the actual amount being even greater in eyes with B-cell lymphoma that sequesters the antibody by antigen binding. Because rituximab preferentially binds to malignant B cells expressing CD20 antigen, it is possible that simple rituximab injection into the vitreous or aqueous humor may be as effective as intralesional treatment for B-cell lymphoma.

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