Outcomes of difluprednate treatment for corneal graft rejection

Nir Sorkin, MD,* Yelin Yang, MD,* Zale Mednick, MD, FRCSC,* Adi Einan-Lifshitz, MD,†
Tanya Trinh, MBBS, FRANZCO,* Gisella Santaella, MD,* Alexandre Telli, MD,*
Clara C. Chan, MD, FACS, FRCSC,* Allan R. Slomovic, MD, FRCSC,* David S. Rootman, MD, FRCSC*

ABSTRACT • RÉSUMÉ

Objective: To evaluate outcomes of difluprednate treatment in penetrating keratoplasty (PK) graft rejection
Design: Retrospective, interventional case series.
Participants: Patients treated with difluprednate for acute endothelial rejection after PK.
Methods: Data were collected on resolution of rejection, treatment regimen used, best spectacle-corrected visual acuity (BSCVA), intraocular pressure (IOP), and side effects. Main outcome measure: rate of rejection resolution. Secondary outcome measures: BSCVA change and side-effect rates.
Results: Thirty-three eyes of 33 patients aged 56.7 ± 17.9 years were included. Twenty-four grafts (72.7%) were high-risk grafts. Complete treatment success was achieved in 19 of 33 grafts (57.6%) over 1.8 ± 1.4 months. Non-high-risk grafts had 100% treatment success rate (9 of 9 grafts). All treatment failures occurred in high-risk grafts, which had a significantly lower treatment success rate of 41.7% (10 of 24 grafts) compared with non-high-risk grafts ($p = 0.004$). Mean BSCVA in the treatment-success group improved from 1.07 ± 0.74 logMAR at the time of rejection to 0.44 ± 0.33 logMAR after treatment ($p = 0.003$). High-dose difluprednate (every 1–3 hours while awake) was used in 93.9% of eyes. IOP elevation and toxic epitheliopathy were each seen in 21.2% of patients. IOP elevation was managed successfully with topical medication and/or difluprednate discontinuation. Epitheliopathy resolved in all cases after completion of difluprednate treatment, except for one case complicated by an infected ulcer.
Conclusions: High-dose difluprednate was effective in treating PK graft rejection, especially in non-high-risk grafts. Adjunct treatment may be required in high-risk grafts. Monitoring for IOP elevation and for toxic epitheliopathy is recommended.

Objectif: Évaluer les résultats de l'administration du difluprednate en cas de rejet du greffon dans la kératoplastie pénétrante (KP).
Nature: Étude rétrospective d'intervention d'une série de cas.
Participants: Patients qui ont reçu le difluprednate en raison d'un rejet endothélial aigu au décours d'une KP.
Méthodes: On a colligé les données sur le taux de préservation du greffon, le schéma thérapeutique utilisé, la meilleure acuité visuelle corrigée (MAVC), la pression intraoculaire (PIO) et les effets indésirables. Parmi les principaux paramètres de mesure, mentionnons le taux de préservation du greffon, tandis que la variation de la MAVC et le taux d'effets indésirables comptaient au nombre des paramètres secondaires.
Résultats: Trente-trois yeux de 33 patients âgés de 56,7 ± 17,9 ans ont été inclus à cette étude. Vingt-quatre greffons (72,7 %) étaient jugés à risque élevé. On a assisté à une réussite complète du traitement dans 19 des 33 greffons (57,6 %) sur une période de 1,8 ± 1,4 mois. Pour les greffons qui n'étaient pas à risque élevé, le taux de réussite du traitement a été de 100 % (9 greffons sur 9). Tous les échecs du traitement se sont produits dans les cas de greffons à risque élevé, chez lesquels le taux de réussite du traitement était significativement moindre: 41,7 % (10 greffons sur 24), comparativement aux greffons qui n'étaient pas à risque élevé ($p = 0,004$). La MAVC moyenne dans le groupe traité avec succès est passée de 1,07 ± 0,74 logMAR au moment du rejet à 0,44 ± 0,33 logMAR après le traitement ($p = 0,003$). Le difluprednate fortement dosé (toutes les 1–3 heures pendant les heures d'éveil) a été administré dans 93,9 % des yeux. Une hausse de la PIO et une épithéliopathie toxique ont chacune été observées chez 21,2 % des patients. L'administration d'un collyre et/ou l'arrêt du difluprednate a permis de juguler efficacement la hausse de la PIO. L'épithéliopathie s'est estompée dans tous les cas, une fois que l'administration du difluprednate a pris fin, sauf dans un cas où un ulcère infecté est venu compliquer le tableau clinique.
Conclusions: Le difluprednate fortement dosé a été efficace dans le traitement du rejet du greffon au décours d'une KP, surtout dans les cas où le greffon n'était pas exposé à un risque élevé. Il pourrait être indispensable de proposer un traitement d'appoint en présence d'un greffon à risque élevé. Il est recommandé de surveiller la hausse de la PIO et de rechercher la présence d'une épithéliopathie toxique.

Immunologic corneal graft rejection is a severe complication after penetrating keratoplasty (PK) with a reported incidence of endothelial graft rejection ranging from 3.5% to 65%, depending on the level of graft vascularity.[1] It causes endothelial cell damage, which can lead to severe endothelial cell loss and graft failure.[2,3] Successful management of a rejection episode requires prompt intervention and aggressive treatment to reverse the rejection process and halt endothelial cell loss as quickly as possible. Corticosteroids are the mainstay of treatment, with topical administration being the treatment of choice and both systemic and periocular administration routes serving as

https://doi.org/10.1016/j.jcjo.2019.07.010
ISSN 0008-4182

adjunct or second-line therapies.[4] Topical steroids are usually administered frequently (up to hourly dosing) and tapered according to clinical response. The most commonly used topical steroidal agents in this scenario are prednisolone and dexamethasone.[4]

Difluprednate ophthalmic emulsion 0.05% (Durezol; Alcon Laboratories, Fort Worth, Tex) is a synthetic prednisolone derivative with high glucocorticoid receptor binding affinity and superior tissue penetration.[5,6] In addition, it has increased bioavailability and dose uniformity resulting from its formulation as an emulsion rather than a suspension.[7] Difluprednate's high potency has been demonstrated in a phase III clinical trial, where difluprednate 0.05% given 4 times daily was found to be noninferior to and possibly more effective than prednisolone acetate 1% given 8 times daily for the treatment of anterior uveitis.[8]

In cases of corneal graft rejection, where aggressive and prompt anti-inflammatory treatment is required to minimize endothelial cell damage, difluprednate's high potency may produce a strong anti-inflammatory effect. The purpose of this study was to evaluate clinical outcomes of difluprednate in the treatment of corneal graft rejection. To the best of our knowledge, this is the first study presenting difluprednate's efficacy in this setting.

Methods

A retrospective medical chart review was performed at Toronto Western Hospital (Toronto, Ont.) on all patients who were treated with difluprednate for acute endothelial rejection after PK. Charts were reviewed starting from difluprednate's approval for use by Health Canada in 2014, and up to 2018. This retrospective interventional case series received Research Ethics Board approval by the University Health Network (Toronto Western Hospital, Toronto, Ont.) and was conducted in compliance with the tenets of the Declaration of Helsinki.

Main Outcome Measures

Efficacy outcomes of interest were resolution of signs and symptoms of rejection together with resolution of edema and clearing of the graft, treatment dosage and duration, change in best spectacle-corrected visual acuity (BSCVA), change in intraocular pressure (IOP), corneal surface status, and rebound of rejection after difluprednate treatment cessation. Patients were also subgrouped according to graft viability after treatment (treatment-success group and treatment-failure group). Comparison of baseline characteristics was made between groups, including demographics, previous graft failure, presence of glaucoma, time between grafting and rejection, and rate of high-risk grafts.

Graft rejection was defined as an endothelial rejection line present in a graft that was previously clear or when there was inflammation (stromal infiltrate, keratic precipitates, cells in the anterior chamber, or ciliary injection) without an endothelial rejection line in a graft that was previously clear. Treatment success was defined as complete resolution of graft edema, together with resolution of rejection signs and symptoms. Graft failure (treatment failure) was defined as nonresolving corneal edema (corneal edema persisting after antirejection treatment), loss of central graft clarity sufficient to compromise vision, or need for a regraft after difluprednate treatment. A high-risk graft was defined in accordance with the Collaborative Corneal Transplantation Studies as a graft with 2 or more quadrants of vascularization, or a graft whose host eye had had a previous rejection episode.[9] Initial difluprednate dosage and taper regimen were determined according to clinical impression of the treating physician.

Statistical Analysis

Data were recorded in Microsoft Excel (2016), and analyzed using SPSS version 23 (SPSS Inc., Chicago, Ill). BSCVA results were converted to logarithm of the minimum angle of resolution (logMAR). Continuous variables were compared within subjects using the Wilcoxon signed-rank test and between subjects using the Mann–Whitney *U* test. Categorical variables were compared using Fisher's exact test. Biserial correlation was used to evaluate association between binary and continuous variables (duration of symptoms and treatment success). All tests were 2-tailed, and the threshold for statistical significance was defined as a p-value <0.05.

Results

Thirty-three eyes of 33 patients with acute endothelial rejection in a PK graft who were treated with difluprednate were included. Average age was 56.7 ± 17.9 years. There were 18 male eyes (54.5%) and 20 right eyes (60.6%). Mean time between corneal grafting and rejection diagnosis was 41.9 ± 63.1 months. Mean duration of symptoms before diagnosis (recorded for 17 patients) was 12.8 ± 9.1 days. Twenty-four grafts (72.7%) were classified as high-risk grafts. Table 1 specifies the surgical indications for PK.

Resolution of Rejection and Graft Survival

Complete resolution of corneal edema and signs of rejection, together with clearing of the graft (treatment success), was achieved in 19 of 33 grafts (57.6%). Mean time to complete resolution was 1.8 ± 1.4 months. Two of the grafts had rejection relapse after difluprednate discontinuation (one at 6

Table 1—Indications for penetrating keratoplasty

Indication	Eyes, n (%)
Failed graft	14 (42.4)
Trauma	6 (18.2)
Keratoconus	5 (15.2)
Herpes simplex / zoster scar	2 (6.1)
Acanthamoeba scar	2 (6.1)
Bacterial ulcer scar	1 (3.0)
Chemical burn	1 (3.0)
Congenital glaucoma	1 (3.0)
Pseudophakic bullous keratopathy	1 (3.0)

Table 2—Comparison of baseline characteristics between treatment-success and treatment-failure groups

Parameter	Treatment Success (N = 19)	Treatment Failure (N = 14)	*p*
Age (years)	53.5 ± 14.7	61.1 ± 21.3	0.177
Male sex (%)	31.6	42.9	0.716
Previous graft failure (%)	31.6	57.1	0.173
History of glaucoma (%)	31.6	50.0	0.472
Time between grafting and rejection (months)	45.3 ± 49.2	36.9 ± 78.7	0.112
High-risk grafts (%)	52.6	100	0.004

months and one at 1.5 months) and were managed successfully with a second treatment course of difluprednate.

Subgrouping of all grafts into high-risk (24 grafts) and non-high-risk (9 grafts) showed that complete resolution was achieved in 10 of 24 eyes (41.7%) in the high-risk group as compared with 9 of 9 eyes (100%) in the non-high-risk group ($p = 0.004$).

A comparison of baseline characteristics was performed between grafts with complete resolution after treatment (19 grafts) and grafts that failed despite treatment (14 grafts). All comparisons were nonsignificant, except for the rate of high-risk grafts that was 52.6% in the treatment-success group versus 100.0% in the treatment-failure group ($p = 0.004$) (Table 2). In the 17 eyes where duration of rejection symptoms before treatment has been recorded, correlation analysis did not find a significant association between duration of symptoms before treatment and treatment success ($p = 0.935$).

Visual Acuity

Mean BSCVA of the entire cohort worsened significantly from 1.11 ± 0.88 logMAR (Snellen equivalent ~20/260) before rejection to 1.34 ± 0.88 logMAR (Snellen equivalent ~20/440) at the time of rejection ($p = 0.025$). After difluprednate treatment, BSCVA improved significantly to 1.00 ± 0.92 logMAR (Snellen equivalent 20/200, $p = 0.002$). This was not significantly different from BSCVA before rejection ($p = 0.418$).

Mean BSCVA in the treatment-success group worsened significantly from 0.78 ± 0.70 logMAR (Snellen equivalent ~20/120) before rejection to 1.07 ± 0.74 logMAR (Snellen equivalent ~20/230) at the time of rejection ($p = 0.033$) and improved significantly after difluprednate treatment to 0.44 ± 0.33 logMAR (Snellen equivalent ~20/50, $p = 0.003$). This was not significantly different from BSCVA before rejection ($p = 0.087$).

Mean BSCVA in the treatment-failure group was 1.56 ± 0.92 logMAR (Snellen equivalent ~20/730) before rejection, 1.71 ± 0.94 logMAR (Snellen equivalent ~20/1025) at the time of rejection ($p = 0.262$), and 2.21 ± 0.77 logMAR (Snellen equivalent 20/3240) after difluprednate cessation ($p = 0.125$). Mean BSCVA after difluprednate cessation was significantly worse than BSCVA before rejection ($p = 0.038$).

Treatment Regimen

Twenty-six of 33 eyes (78.8%) were receiving prophylactic topical steroidal treatment before rejection with prednisolone acetate, dexamethasone, fluorometholone, or loteprednol. Dosage ranged from once daily to 4 times daily. One patient was receiving difluprednate twice daily as topical prophylactic treatment. None of the patients received systemic steroids while being prescribed difluprednate.

Difluprednate was used as first-line rejection treatment in 27 of 33 eyes (81.8%). The remaining 6 eyes (18.2%) were initially treated with hourly topical steroids other than difluprednate (prednisolone acetate in 5 eyes and loteprednol in 1 eye) and were switched to difluprednate due to insufficient clinical improvement (treatment time before difluprednate switch ranged between 5 and 21 days). Three of those eyes (50%) had treatment success after switching to difluprednate, with complete resolution of rejection.

Initial difluprednate dosing was hourly while awake in 18 eyes (54.5%), every 2 hours while awake in 11 eyes (33.3%), and every 3 hours while awake in 2 eyes (6.1%). The remaining 2 eyes (6.1%) were managed initially with 4 times daily and 5 times daily dosing. Tapering of difluprednate treatment was done according to the clinical response. None of the eyes received any additional antirejection medication during difluprednate treatment. In eyes where rejection resolved completely, the mean duration of difluprednate treatment was 13.8 ± 13.4 weeks. Figure 1 shows the mean difluprednate tapering regimen during the first 3 months in eyes where complete rejection resolution was achieved.

Safety

Seven eyes (21.2%) had IOP elevation during difluprednate treatment, ranging between 4 and 11 mm Hg. Only 1 eye had a maximal IOP >30 mm Hg (value of 35 mm Hg), with the remaining 6 eyes having maximal IOP values of 20−28 mm Hg. Median timing of IOP elevation was 38.5 ± 64.5 days (range 14−183 days), with only 4 eyes (12.1%) having IOP elevation during the first treatment month. Difluprednate dosage at the time of IOP elevation ranged from twice daily to every hour while awake, with 2 eyes only having IOP elevated while on high-dose treatment (one was on every hour while awake dosing and one on every 2 hours while awake dosing).

Topical IOP-lowering treatment was added in 6 of 7 eyes. Also, difluprednate was switched to a different topical steroid in 6 of 7 eyes. None of the patients required surgical intervention to manage their IOP elevation. The rate of IOP elevation in previously diagnosed glaucoma patients was 23% (3 out of 13 glaucoma patients).

Seven eyes (21.2%) developed toxic epitheliopathy during difluprednate treatment. Epitheliopathy was mild in 4 eyes (superficial punctate keratitis) and moderate in 3 eyes (small epithelial defect). Epitheliopathy resolved completely in 6 eyes after discontinuation of difluprednate. One eye treated with a bandage contact lens for a small epithelial defect

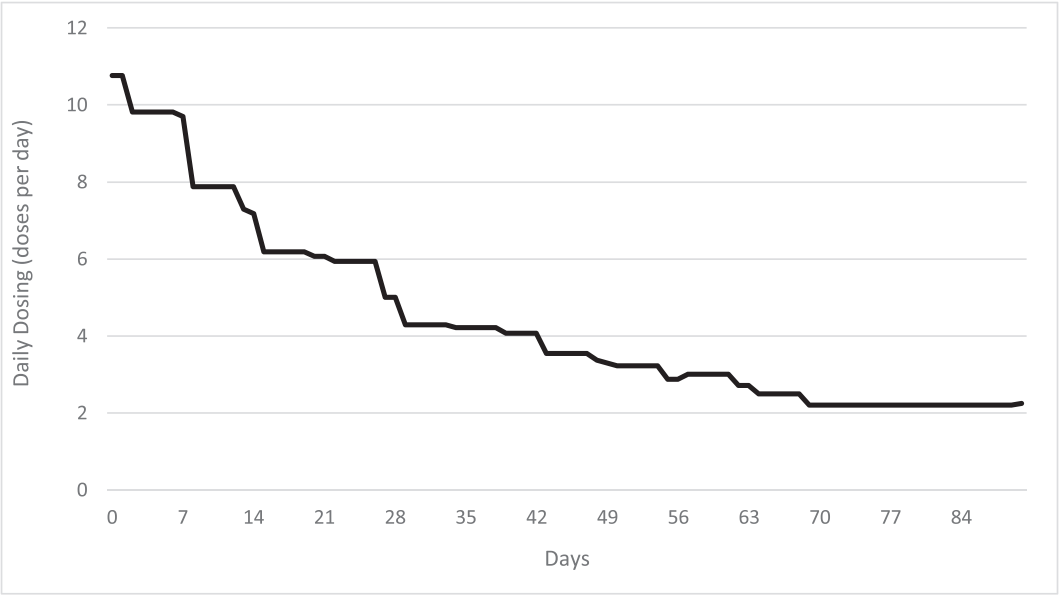

Fig. 1 — —Mean difluprednate tapering regimen in the treatment-success group (n = 19) during the first 90 treatment days.

developed an infected corneal ulcer, which resolved with fortified topical antibiotics, but incited a repeat rejection episode with subsequent graft failure.

Discussion

The present study is the first to evaluate outcomes of corneal graft rejection managed with topical difluprednate. Due to its high potency, difluprednate can induce a strong anti-inflammatory effect that may be critical in cases of endothelial graft rejection. This effect can be maximized using a high-dose regimen.

Rates of complete rejection reversal and graft clearing in the published literature vary between 42% and 92%.[9–13] The overall treatment success rate in the present study was 57.6%, but subgrouping showed that 100% of the non-high-risk grafts had complete treatment success. Additionally, we found that 100% of the failed grafts were high-risk grafts. These findings suggest that difluprednate is highly effective in grafts that are not high risk, and could be considered as a single first-line agent in such cases. However, in cases of high-risk grafts, adjunct treatments such as systemic or periocular steroids should be considered. A small group of patients in our study (6 eyes) received difluprednate as second-line rejection treatment after failure of other topical steroids. A success rate of 50% in this group suggests that difluprednate might have a place as second-line treatment in cases of first-line failure with other topical agents, possibly combined with systemic or periocular therapy.

The vast majority of eyes in this study (93.9%) initially received high-dose (every 1−3 hours while awake) difluprednate, similar to topical treatment protocols employed with other steroidal topical agents for graft rejection. This enables a maximal anti-inflammatory effect during the early treatment period. The safety profile of high-dose difluprednate was reasonable, with IOP elevation seen in 21% of the eyes. This is similar to rates of steroid response to other topical steroids in the general population[14] and is also comparable to a 21% IOP elevation rate reported by Schallhorn et al. in uveitic cystoid macular edema patients treated with low-dose difluprednate for 3 months.[15] A substantial portion of IOP elevation in our study was delayed, with only 4 eyes (12%) experiencing IOP rise during the first treatment month. These findings are again comparable to an 11% IOP elevation rate after 1 month of difluprednate treatment found in the study of Schallhorn et al.[15] These findings, along with the fact that only 2 eyes in our study experienced IOP elevation while under high-dose difluprednate, suggest that IOP response to difluprednate is more a function of treatment duration rather than dosage.

Mild toxic epitheliopathy from difluprednate use has been previously reported with low dosing for anterior uveitis and postoperative inflammation at rates ranging between 3.6% and 16.0%.[8,16,17] These rates were also found to be higher in difluprednate-treated eyes when compared with both placebo and prednisolone acetate. In the current study, epitheliopathy rates were higher (21.2%), possibly due to higher difluprednate dosage and because treatment was given over a PK graft surface. These findings corroborate our clinical impression that high-dose difluprednate use is associated with increased rates of toxic epitheliopathy, and therefore close ocular surface monitoring is suggested—especially in patients with a poor ocular surface. Given the above-described rates of epitheliopathy and IOP elevation, together with difluprednate's success rate in cases of high-risk corneas, which seems comparable to that of other topical steroids, further evaluation of difluprednate's role in such cases is warranted. Prospective trials comparing difluprednate and other topical steroids could

help establish the preferred topical steroid to be used in various settings of corneal graft rejection.

This study is limited by its retrospective nature and cohort size. Also, data on duration of rejection symptoms before diagnosis were limited and did not enable its incorporation into the analysis. Nevertheless, this is the first report on the outcomes of difluprednate treatment in PK graft rejection, as well as the first report on the use of a high-dose difluprednate regimen.

In conclusion, high-dose difluprednate was effective in treating endothelial PK graft rejection, especially in non-high-risk grafts. Adjunct treatment may be required in high-risk grafts. Monitoring for IOP elevation and for toxic epitheliopathy is recommended. Given difluprednate's efficacy and safety profile, the role of difluprednate in the management of corneal graft rejection should be further evaluated.

References

1. Panda A, Vanathi M, Kumar A, Dash Y, Priya S. Corneal graft rejection. *Surv Ophthalmol*. 2007;52:375–96.
2. Wilson SE, Kaufman HE. Graft failure after penetrating keratoplasty. Surv Ophthalmol. n.d.;34:325–56.
3. Vail A, Gore SM, Bradley BA, Easty DL, Rogers CA. Corneal graft survival and visual outcome: a multicenter study. Corneal Transplant Follow-up Study Collaborators. *Ophthalmology*. 1994;101:120–7.
4. Kharod-Dholakia B, Randleman JB, Bromley JG, Stulting RD. Prevention and treatment of corneal graft rejection. *Cornea*. 2015;34:609–14.
5. Bikowski J, Pillai R, Shroot B. The position not the presence of the halogen in corticosteroids influences potency and side effects. *J Drugs Dermatol*. 2006;5:125–30.
6. Gysler A, Lange K, Korting HC, Schäfer-Korting M. Prednicarbate biotransformation in human foreskin keratinocytes and fibroblasts. *Pharm Res*. 1997;14:793–7.
7. Stringer W, Bryant R. Dose uniformity of topical corticosteroid preparations: difluprednate ophthalmic emulsion 0.05% versus branded and generic prednisolone acetate ophthalmic suspension 1%. *Clin Ophthalmol*. 2010;4:1119–24.
8. Sheppard JD, Foster CS, Toyos MM, et al. Difluprednate 0.05% versus prednisolone acetate 1% for endogenous anterior uveitis: pooled efficacy analysis of two phase 3 studies. *Ocul Immunol Inflamm*. 2019;27:484–96.
9. The Collaborative Corneal Transplantation Studies (CCTS): effectiveness of histocompatibility matching in high-risk corneal transplantation. *Arch Ophthalmol*. 1992;110:1392–403.
10. Sangwan VS, Ramamurthy B, Shah U, Garg P, Sridhar MS, Rao GN. Outcome of corneal transplant rejection: a 10-year study. *Clin Exp Ophthalmol*. 2005;33:623–7.
11. Wagoner MD, Ba-Abbad R, Sutphin JE, Zimmerman MB. Corneal transplant survival after onset of severe endothelial rejection. *Ophthalmology*. 2007;114:1630–7.
12. Hudde T, Minassian DC, Larkin DFP. Randomised controlled trial of corticosteroid regimens in endothelial corneal allograft rejection. *Br J Ophthalmol*. 1999;83:1348–52.
13. Naacke HG, Borderie VM, Bourcier T, Touzeau O, Moldovan M, Laroche L. Outcome of corneal transplantation rejection. *Cornea*. 2001;20:350–3.
14. Jones R, Rhee DJ. Corticosteroid-induced ocular hypertension and glaucoma: a brief review and update of the literature. *Curr Opin Ophthalmol*. 2006;17:163–7.
15. Schallhorn JM, Niemeyer KM, Browne EN, Chhetri P, Acharya NR. Difluprednate for the treatment of uveitic cystoid macular edema. *Am J Ophthalmol*. 2018;191:14–22.
16. Korenfeld MS, Silverstein SM, Cooke DL, Vogel R, Crockett RS. Difluprednate ophthalmic emulsion 0.05% for postoperative inflammation and pain. *J Cataract Refract Surg*. 2009;35:26–34.
17. Foster CS, DaVanzo R, Flynn TE, McLeod K, Vogel R, Crockett RS. Durezol (difluprednate ophthalmic emulsion 0.05%) compared with Pred Forte 1% ophthalmic suspension in the treatment of endogenous anterior uveitis. *J Ocul Pharmacol Ther*. 2010;26:475–83.

Footnotes and Disclosure

The authors have no proprietary or commercial interest in any materials discussed in this article.

From the *Department of Ophthalmology and Vision Sciences, University of Toronto, Toronto, Ont.; †Ophthalmology Department, Assaf Harofeh Medical Center, Sackler Faculty of Medicine, Tel Aviv University, Tel Aviv, Israel.

Originally received Feb. 26, 2019. Final revision Jun. 16, 2019. Accepted Jul. 12, 2019.

Correspondence to Nir Sorkin, MD, Department of Ophthalmology and Vision Sciences, Toronto Western Hospital, 399 Bathurst Street, 6th Floor East Wing, Reception 1, Toronto, Ont. M5T 2S8. nir.sorkin@gmail.com