Endothelial cells analysis after intraoperative mitomycin-C adjuvant pterygium simple excision surgery: a pilot study

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Abstract

Purpose: To study the effects of mitomycin-C (MMC) on the corneal endothelium after pterygium surgery.

Methods: Prospective, interventional, nonrandomized, observer-masked study. Twenty patients (20 eyes) with primary pterygium underwent surgery with pterygium simple excision followed by 0.02% MMC application for 1 minute. Preoperative, 10-day, 1-month and 3-month postoperative specular microscopy were performed. Mean cell density, coefficient of variation of mean cell area and percentage of hexagonal cells were measured.

Results: Mean endothelial cell densities before and 3 months after surgery were 2,240.37 SD 347.92 cells/mm² (range: 1,020 to 2,703) and 2,283.86 SD 425.13 cells/mm² (range: 1,001 to 2,837), respectively. Mean coefficient of variation before and 3 months after surgery were 32.63 SD 4.65 (range: 22 to 48) and 33.00 SD 4.75 (range: 23 to 51), respectively. Mean percentage of hexagonal cells before and 3 months after surgery were 54.58 SD 6.60 (range: 46 to 71) and 55.26 SD 8.89 (range: 38 to 76), respectively. No statistically significant differences were found in any of the parameters studied when compared to the preoperative values.

Conclusions: A single intraoperative application of 0.02% MMC for 1 minute during pterygium simple excision surgery does not seem to cause substantial changes in corneal endothelial cell density at 3 months.

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Introduction

Pterygium is a degenerative process characterized by a fibrovascular growth of the conjunctiva towards the cornea which, in severe cases, could produce significantly diminished vision. Pterygium is more prevalent in populations living close to the Equator. The main indications for the surgical excision of pterygium are diminished visual acuity, ocular irritation which does not diminish with lubrication, ocular motility restriction, progressive growth towards the visual axis, problems in the use of contact lenses, refractive surgery and aesthetic alterations.

Numerous surgical techniques have been described as methods for treating primary pterygium, such as simple excision with denuded sclera, simple excision with application on the denuded sclera of mitomycin C (MMC) in different dosages and concentrations and the use of conjunctival self-grafts after pterygium excision. However, post-op relapses are a severe problem. In order to reduce the relapse rate, coadjuvant therapies have been utilized in recent decades with varying degrees of success, such as beta-irradiation, MMC, 5-flouracyl and thiotepa.

The use of MMC as an adjuvant therapy for pterygium surgery was described by Kunitomo and Mori in 1963 and in the United States by Singh et al in 1988. MMC is an antibiotic-antineoplastic agent that selectively inhibit the synthesis of DNA, cellular RNA and proteins, the action mechanism of MMC in the prophylaxis of recurring pterygium is explained by the inhibition of the fibroblastic proliferation at the episcleral level. Post-op administration of topical MMC in concentrations between 0.02% (0.2 mg/ml) and 0.1% (1 mg/ml) reduces the pterygium recurrence rates to a range between 2% and 11%.

The objective of this study is to perform a prospective short term analysis by means of mirror microscopy of the possible induced toxicity on the corneal endothelium by the intra-op administration of 0.02% MMC during one minute in simple pterygium excision.

Subjects, material and methods

Patients

Twenty eyes of 20 patients with primary pterygium were included in this non-randomized, interventional and
prospective study with masked observer. All the participants signed the informed consent. The inclusion criteria were: a) primary pterygium with flesh and active pterygium invading the cornea beyond 3 mm; b) no previous surgery or traumatism in the affected eye; c) not being a contact lens user, and d) being free of ocular and systemic diseases which could alter the morphology of the corneal endothelium such as diabetes and connective tissue diseases.

The patients underwent a complete ophthalmological exploration: LogMAR Best corrected visual acuity (BCVA), refraction, biomicroscopy, applanation tonometry and orthaphthalmoscopy. A simple pterygium excision was performed with intraop application of MMC 0.2 mg/ml (0.02%) directly on the scleral bed during one minute utilizing a microsurgery swab (Alconcusi, Barcelona). At all times contact of the swab with a peripheral cornea was avoided and subsequently the ocular surface was abundantly irrigated with a balanced serum solution. After surgery, the patients were assessed after one and 10 days, 1 and 3 months. In the last three evaluations their BCVA, refraction, conjunctival injection, epithelial corneal defects and pterygium recurrence were assessed. Recurrence was defined as a post-op growth of fibrovascular tissue through the limbus towards the cornea.

Endothelial cell analysis

Contact-free mirror microscopy was performed on the central cornea with a Topcon SP-3000P microscope. In the pre-op and post-op assessments after 10 days, 1 month and 3 months, a masked observer (JBH) took three images of the central cornea of which only the clearest and best focused image was filed for analysis. The microscope automatically evaluated the density of endothelial cells, the variation coefficient (an objective polymegatism measure) and the cellular hexagonality percentage (pleomorphism index). The same masked observer carried out a manual analysis of the endothelial cells. At least 75 cells with well-defined edges were manually counted within a defined square of the image, corresponding to 0.03 mm² of the actual cornea.

Statistical analysis

All the data were expressed as mean and standard deviation (SD) values. The differences between the pre- and post-op data were statistically evaluated by means of the t for Student test and the p < 0.01 values were considered significant for correcting the multiple comparison effects (Bonferroni correction). The statistical analysis was processed by means of SPSS 14.0 for Windows (SPSS Inc, Chicago, IL, USA).

Results

Of the 20 patients included in the study, 12 (60%) were male and 8 (40%) female. The mean age of these patients was of 62 SD 15.08 years (range: 37 to 83 years). Eleven right eyes and nine left eyes were assessed. In 18 eyes the pterygium was nasal and in two, nasal and temporal.

No statistically significant differences were found (p = 0.53) between pre-op BCVA (0.251 SD 0.281) and post-op BCVA (0.253 SD 0.349) at month three after surgery. All the patients exhibited in the immediate post-op (1-10 days) discomfort, tearing and photophobia in relation to the irritation caused by the suture. One month after surgery, six patients exhibited slight-moderate conjunctival injection and four of these also exhibited epithelial corneal defects. Three months after surgery they were free of symptoms, without conjunctival injection or epithelial corneal defects in any case. No important post of complications arose such as corneal or scleral necrosis. Two patients (10%) exhibited pterygium recurrence in the post-op period comprised between 1-3 months.

Table 1 shows the effect of intra-op MMC on the corneal endothelium. The mean endothelial cellular density before and after surgery up to 10 days, one and 3 months was or 2.240.37 SD 347.92 cells/mm² (range: 1.020 to 2.703), 2.262.05 SD 369.15 cells/mm² (range: 1.030 to 2.795), 2.260.28 SD 369.69 cells/mm² (range: 981 to 2.720) and 2.283.86 SD 425.13 cells/mm² (range: 1.001 to 2.837), respectively. The mean variation coefficients before and after surgery, after 10 days, one month and 3 months were: 32.63 SD 4.65 (range: 22 to 48), 32.89 SD 5.50 (range: 23 to 51), 32.61 SD 4.98 (range: 22 to 50) and 33.00 SD 4.75 (range: 23 to 51) respectively. The mean cellular hexagonality percentages before and after surgery, after 10 days, one month and 3 months were 54.58 SD 6.60 (range: 46 to 71), 53.59 SD 10.33 (range: 30 to 68), 55.00 SD 9.10 (range: 43 to 74) and 55.26 SD 8.89 (range: 38 to 76), respectively. The differences were not statistically significant in any of the analyzed variables or in the post-op assessment periods.

Discussion

This study has demonstrated that intraop administration of 0.02% MMC during 1 minute in simple pterygium excision

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<th>Table 1 – Effect of intraop mitomycin C on the corneal endothelium. All data in mean and standard deviation (SD)</th>
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<tr>
<td><strong>n = 20</strong></td>
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<td>Preop</td>
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does not produce toxic effects at the corneal endothelial level. No statistically significant differences were found between the pre-op and post-op values in all the assessed intervals (10 days, 1 month, 3 months) on the analyzed endothelial morphometric parameters such as corneal endothelium cell density, cell size variation coefficient and cell hexagonality percentage. Therefore, it can be said that intraop administration of MMC as a coadjuvating treatment to prevent the recurrence of pterygium does not have significant toxic effects on the corneal endothelium in the short term, after a 3-month follow-up period.

This study has some limitations related to the small sample size (n = 20) and the broad range of pre-op endothelial cell density. However, it has allowed us to determine the number of patients needed to attain statistical significance for an 80% study power and a 5% alpha error assuming that the differences found between the pre-op and post-op values as regards corneal endothelial cell density would maintain the same trend. Accordingly, these results are supported by a larger study carried out in 51 eyes of 46 patients with primary pterygium, recently published by our group22. Another possible limitation is the short follow-up period (3 months) of our study, taking into account the cumulative and delayed toxic effects of MMC23. Therefore, it would be convenient to carry out additional studies about the toxic effect of MMC analyzing the long-term results.

Recently, several studies have researched the toxic effect of MMC on the corneal endothelium in pterygium surgery22,24-26. Even though the intraop application of MMC in simple pterygium excision is effective to prevent recurrences, its toxic effect varies with the dosage and time of application. The optimum application protocol has not yet been established15-21. Avisar et al24 reported an immediate and sustained effect for a 90-day follow-up period on the endothelial cell density after the intraop application of MMC at 0.02% during five minutes on the bare sclera after simple primary pterygium excision. This was not demonstrated by the authors25 when the same MMC dosage was applied before removing the pterygium head. Bahar et al26 also demonstrated that the intraop application of 0.02% MMC during two minutes had toxic effects on the corneal endothelium in recurring pterygium surgery. In our study, carried out on primary pterygium surgery, we applied MMC on the scleral bed in the same concentration as regards corneal endothelial cell density would maintain the same trend. Accordingly, these results are supported by a larger study carried out in 51 eyes of 46 patients with primary pterygium, recently published by our group22. Another possible limitation is the short follow-up period (3 months) of our study, taking into account the cumulative and delayed toxic effects of MMC23. Therefore, it would be convenient to carry out additional studies about the toxic effect of MMC analyzing the long-term results.

In conclusion, a single intraop application of 0.02% MMC during one minute after a simple pterygium excision does not seem to cause substantial changes on the corneal endothelial cell density after a three-month follow-up period.

REFERENCES


