No effects of mitomycin-C in primary trabeculectomies in Sweden

Marcelo Ayala

Abstract
Aim: To evaluate the results of a long-term follow-up after two different types of surgical techniques: trabeculectomy with or without mitomycin-C.

Materials and methods: This study is a retrospective chart review of patients operated on with a primary trabeculectomy at the Eye Department of the Skaraborg Hospital, Skövde, Sweden. Complete success was defined as intraocular pressure ≤18 mmHg (criterion 1) or intraocular pressure reduction ≥30% (criterion 2) without eye drops postoperatively. Qualified success was defined using the same criteria (1 and 2), but patients were treated or untreated with eye drops.

Results: A total of 167 patients were included in this retrospective study, 83 patients in the no-mitomycin-C group and 84 patients in the mitomycin-C-treated group. No significant difference was found in intraocular pressure reduction between the mitomycin-C and no-mitomycin-C group (t-test, p = 0.19). Complete success using criterion 1 was 66.2% in no-mitomycin-C and 62.8% in mitomycin-C (p = 0.88); success using criterion 2 was 76.6% in the no-mitomycin-C and 64.2% in the mitomycin-C group (p = 0.21). Qualified success using criterion 1 was 71.4% in the no-mitomycin-C and 74.4% in the mitomycin-C group (p = 0.84); success using criterion 2 was 80.0% in the no-mitomycin-C and 84.4% in the mitomycin-C group. All included patients were born in Sweden.

Conclusion: Mitomycin-C seems to add no benefits to intraocular pressure reduction after primary trabeculectomies in a Swedish population.

Keywords
Mitomycin-C, glaucoma, surgery, Sweden

Date received: 22 February 2018; accepted: 17 May 2018

Introduction
Glaucoma is the second most common age-related eye disease in the world.1 Preliminary calculations showed that about 60 million people over 40 are affected around the world.2 The disease affects mainly older people, and an estimated 1%–2% of Swedes will suffer from glaucoma at some point. This disease is the second most common cause of blindness in the world.3

Glaucoma is a neurodegenerative disorder characterized by the loss of retinal ganglion cells, leading to atrophy of the optic nerve, thus progressive visual field effect/visual impairment. The disease is usually caused by increased pressure in the eye, but patients with normal intraocular pressure (IOP) can also suffer from glaucoma. There is no cure for glaucoma, and treatment is therefore intended to slow down its progress by lowering eye pressure.4 The IOP can be lowered using eye drops laser or surgery. Still, it is unknown whether any of the latter methods are better than others.5

There are different methods for operating on glaucoma. Trabeculectomy is still the ‘gold standard’ among the different surgical methods.6 There is a wide variety of methods, including trabeculectomy without additional treatment, trabeculectomy plus mitomycin-C (MMC), 5-fluorouracil (5-FU) and so on. MMC’s alkylating properties inhibit DNA replication, which led to its use first as an anti-cancer drug. MMC is an agent that prevents scarring by inhibiting the multiplication of cells that produce scar tissue. Other types

Eye Department, Skaraborg Hospital Skövde, Sahlgrenska Academy, University of Gothenburg and Karolinska Institute, Skövde, Sweden

Corresponding author:
Marcelo Ayala, Eye Department, Skaraborg Hospital Skövde, Sahlgrenska Academy, University of Gothenburg and Karolinska Institute, SE-541 85 Skövde, Sweden.
Email: marcelo.ayala@vgregion.se; marcelo.ayala@ki.se
of glaucoma surgery include deep sclerectomy, canaloplasty and various types of shunt procedures. Comparisons among studies are difficult to perform due to variation between different patient populations, diverse glaucoma stages, different surgeons, other therapies and so on. The purpose of this study was to evaluate the results of a 2-year follow-up after adding MMC to trabeculectomy.

Materials and methods

This is a retrospective case–control study with a non-concurrent comparison group; we performed retrospective data collection with different time periods for the two groups (chart review). The method was chosen due to the fact that between 2004 and 2007, nearly all glaucoma patients were operated by means of trabeculectomy without MMC. Between 2008 and 2009, different techniques were used, such as trabeculectomy with implants, deep sclerectomy and so on. From 2009 to 2018, nearly all patients were operated on by means of trabeculectomy with MMC. Inclusion criteria included patients who underwent primary trabeculectomy with or without MMC at the Department of Ophthalmology, Skaraborg Hospital (SKAS), Skövde, Sweden.

Included patients were operated on between 2004 and 2007 for the group without MMC and between 2009 and 2012 for the group with MMC. All patients were followed up for at least 2 years after surgery. Included patients were suffering from normal tension glaucoma (NTG), secondary glaucoma, primary open angle glaucoma (POAG) or exfoliation glaucoma. Exclusion criteria included patients operated on with a technique other than trabeculectomy, patients operated on before with a trabeculectomy and patients suffering from diseases other than glaucoma. The following variable was examined: eye pressure after surgery. Success or failure was classified according to the World Glaucoma Association (WGA) Guidelines on Design and Reporting of Glaucoma Surgical Trials. The whole study followed the recommendations of the WGA. The study was designed according to the recommendations of the STROBE guidelines (www.strobe-statement.org).

Definitions

Definitions used in this study were those stipulated by the WGA. Complete failure was defined as loss of light perception attributable to glaucoma or the necessity for further glaucoma intervention. Certain postoperative surgical adjustments, such as flap suturelysis, suture release, needling and so on, had not been recorded as evidence of failure. Successful result includes eye pressure \(\leq 18\) mm Hg (criterion 1) or pressure reduction \(\geq 30\%\) (criterion 2) from the primary status that had been measured for at least two time points. Complete success was defined as IOP \(\leq 18\) mm Hg (criterion 1) or pressure reduction \(\geq 30\%\) (criterion 2) without medication. Qualified success was defined as IOP \(\leq 21\) mm Hg (criterion 1) or pressure reduction \(\geq 30\%\) (criterion 2) with or without medication. Criteria 1 and 2 were chosen according to the suggestions of the WGA for moderate glaucoma.

All patients underwent detailed ophthalmological examinations before the operation. The best-corrected visual acuity was measured using Snellen charts. IOP was measured at least twice on different days using an adequately calibrated Goldmann Applanation Tonometer (GAT). Baseline IOP before surgery was defined as IOP on the patient’s full medical regimen just before surgery. Gonioscopy was performed with the patient sitting at the slit-lamp using a Zeiss-type four mirror gonioprism to examine the anterior chamber angle for neovascularization, pigmentation and grade of depth. Just one eye per patient was included in the study; when both eyes were operated on, one eye was chosen at random. A dilated fundus examination was performed. After pupil dilation, the optic nerve and posterior pole were examined at the slit lamp using a Volk 90 dioptre lens.

Preoperative and postoperative medications were recorded as the number of medications and not as the number of bottles, as recommended by the WGA.

Surgical technique. All patients were operated on under local anaesthesia. A conjunctival peritomy was created to expose the superior bare sclera. This peritomy was fornix-based. Dissection of the subconjunctival space to provide a pathway for the aqueous flow was then performed.

A rectangular partial thickness scleral flap was created in the superior sclera, hinged at the limbus. The scleral flap was dissected forward until the bluish grey zone at the limbus was exposed. A sponge embedded in MMC 0.4 mg/mL was then placed on the bare sclera for 3 min. The sponge was removed afterwards, followed by generous irrigation of the subconjunctival space. An ostomy was then created underneath the scleral flap using a Kelly punch device. A peripheral iridectomy was created using forceps to pull the peripheral iris through the ostomy site and scissors to create the iridectomy. Two sutures (10-0 nylon) were then used to close the scleral flap. The surgical technique was similar in the patients operated without MMC, but in this case, MMC was not used. The flap was closed tightly enough to allow the anterior chamber to remain formed. The conjunctival peritomy was then closed using absorbable sutures. Five different surgeons performed the surgery. Four of them were the same for both groups: with or without MMC. One surgeon differed between the groups. However, all the surgeons used the same surgical technique. The author is not one of the surgeons.

Surgical follow-up

After glaucoma filtration surgery, the patient was seen on the first post-operative day. All patients were treated with dexamethasone 1 mg/mL eye-drops (Isopto-Maxidex; Alcon Laboratories, Fort Worth, Texas, USA) five times daily for
Table 1. Baseline characteristics of the patients studied.

|                          | Group 1 (no MMC) | Group 2 (MMC) | Test         | p     |
|--------------------------|------------------|---------------|--------------|-------|
| Gender (F/M)             | 49/34            | 44/40         | Chi-square ($\chi^2$) | 0.11  |
| Diagnosis (NTG/sec/POAG/Exf.) | 2/3/25/53       | 2/4/20/58     | Chi-square ($\chi^2$) | 0.86  |
| Age                      | $\bar{x} = 74.86 \pm 10.46.$ | $\bar{x} = 73.14 \pm 9.16.$ | t-test       | 0.27  |
| Number of medications before surgery | Range = 65–89   | Range = 64–88 |              |       |
| IOP before surgery (mmHg) | $\bar{x} = 3.72 \pm 0.45$ | $\bar{x} = 3.58 \pm 0.59$ | t-test       | 0.10  |
| Phakia/pseudophakia      | 63/20            | 56/28         | Chi-square ($\chi^2$) | 0.10  |
| Visual fields mean deviation (MD) (dB) | 10.57 ± 2.34    | 10.24 ± 2.22  | t-test       | 0.89  |
| Optic nerve cup/disc ratio | 0.88 ± 0.1     | 0.87 ± 0.13   | t-test       | 0.46  |
| Glaucoma stage           |                 |               |              |       |
| (European)               | Early (MD $\leq$ −6 dB) (N/%) | 6 (7.2%)     | 7 (8.3%)     | Chi-square ($\chi^2$) | 0.78  |
|                          | Moderate (MD $\geq$ −6 < −12 dB) (N/%) | 69 (83.1%)   | 71 (84.6%)   | Chi-square ($\chi^2$) | 0.86  |
|                          | Advanced (MD $\geq$ −12 dB) (N/%) | 8 (9.7%)      | 6 (7.1%)     | Chi-square ($\chi^2$) | 0.59  |

MMC: mitomycin-C; IOP: intraocular pressure.

4 weeks post-operatively. A topical antibiotic was prescribed to be used four times daily for the first week. No other kind of topical medication was used (5-FU, MMC, etc.). Depending on the patient’s clinical state and IOP, the follow-up interval varied. A typical routine was to see the patient 1 week postoperatively and then every 1–2 weeks afterwards for the first 2 months. Then, the patients were checked every 4 months.

Complications were recorded according to the recommendations of the WGA. Hypotony was defined as IOP $\leq 5$ mmHg. Cataract formation was defined as reduction in the optical clarity of the natural lens producing sufficient visual disturbance to require surgery. Endophthalmitis was defined as infection of the globe contents that, even with prompt aggressive treatment, often results in substantial loss of visual function.

Choroidal detachment was defined as detachment of the choroid layer visualized after pupil dilation examined at the slit lamp using a Volk 90 dioptre lens. The diagnosis was confirmed using an ultrasound device (Quantel Medical, Clermont-Ferrand, France).

Statistics. The study follows the Guidelines on Design and Reporting of Glaucoma Surgical Trials (WGA). The guidelines recommend the use of Kaplan–Meier survival analysis than a simple comparison between groups at a given follow-up time. The two groups, that is, the two different types of surgical techniques (trabeculectomy with or without MMC) were compared using the log-rank test (Mantel–Cox). Kaplan–Meier curves and log-rank test were performed using the SPSS software (IBM, New Orchard Road Armonk, New York, USA).

Sample size was also estimated according to the recommendations of the WGA. The guidelines recommend the use of Power and Sample Size Calculation (PS), a useful sample size programme available free from the Vanderbilt University (http://biostat.mc.vanderbilt.edu). Calculation of sample size assumes an alfa error = 0.05 and a statistical power of 80%. The study will include one control per experimental subject and an accrual interval of 24 months with no additional follow-up after the accrual interval. The median survival time on the control treatment was set to 18 months. If the true hazard ratio (relative risk) of control subjects relative to experimental subjects is 0.5, the study will need to include 77 experimental subjects and 77 control subjects to be able to reject the null hypothesis. The null hypothesis is that the experimental and control survival curves are equal with a power of 80%. The alfa error associated with this test of the null hypothesis is 0.05.

The study followed the Tenets of the Helsinki Declaration. The study was approved by the Gothenburg Ethical Committee (DN: 1050-13).

Results

A total of 167 patients were included in this retrospective study: 83 in the no-MMC group and 84 in the MMC-treated group. All included patients were White individuals born in Sweden and their parents also were born in Sweden. In four cases, parents were born in Finland. No Africans, Asian and other non-White patients were included.

No significant differences were found between the groups at baseline regarding gender ($p = 0.11$), diagnosis ($p = 0.86$), age ($p = 0.27$), number of medications ($p = 0.10$), IOP before surgery ($p = 0.35$), frequency of phakia/pseudophakia ($p = 0.10$) or visual field damage ($p = 0.89$) (Table 1).

Regarding the IOP reducing effect of trabeculectomy, in the no-MMC group, IOP decreased from preoperative average $28.78–13.91$ mmHg (difference $= 14.87$ mmHg) postoperatively. The IOP reduction was significant (t-test; $p \leq 0.0001$). In the MMC group, IOP decreased from preoperative average $27.80–12.72$ mmHg (difference $= 15.07$ mmHg) postoperatively. The IOP reduction was significant (t-test; $p \leq 0.0001$). The IOP reduction comparing both groups (no MMC vs MMC) showed to be no significant (t-test; $p = 0.19$).
Complete failure was $n = 4$ (4.8%) in the no-MMC group and $n = 5$ (6%) in the MMC group. The difference was statistically insignificant ($\chi^2; p = 0.85$). In both groups, all patients were classified as ‘failure’ if new glaucoma surgical intervention was needed within 2 years after initial surgery.

Complete and qualified success did not differ significantly between the no-MMC and MMC group. The results are presented in Figures 1–4 and summarized in Table 2.

Before surgery, all patients ($n = 167$) were being treated with medications. The average number of medications was $3.65 \pm 0.52$: $3.72 \pm 0.45$ in the no-MMC group and $3.58 \pm 0.59$ in the MMC group. At the end of the study, $n = 130$ (78%) patients were not taking any medication. A total of $n = 70$ (84%) of them were in the No-MMC group and $n = 60$ (71%) in the MMC group. The difference was not significant ($\chi^2$, $p = 0.06$). The average number of medications after surgery was $1.92 \pm 0.74$. The number of medications was $1.68 \pm 0.67$ in the no-MMC group and $2.16 \pm 0.82$ in the MMC group. The difference was statistically significant (t-test; $p = 0.04$).

A significant majority of the operations were performed by the same surgeon: $n = 48$ (59%) in the no-MMC group and $n = 50$ (60%) in the MMC group. Another surgeon performed $n = 18$ (22%) in the no-MMC and $n = 17$ (20%) in the MMC group. The rest of the operations (around 20%) were performed by three different surgeons (two of them included in both the no-MMC and MMC groups). All surgeons were senior consultants. No significant difference was found among the various surgeons regarding IOP reduction after surgery in the MMC group (ANOVA: $F = 0.87, p = 0.48$). IOP reduction after surgery was measured as a percentage. Nor was any significant difference found in the...
no-MMC group among different surgeons regarding IOP reduction after surgery (ANOVA: F = 0.22, p = 0.88).

Complications were graded according to the WGA recommendations. Hypotony, defined as IOP $\leq 5$ mmHg, was of 24.1% in the no MMC meanwhile in the MMC group the frequency was 46.4%. The difference was statistical significant (chi-square, $p = 0.0002$). Hypotony was probably induced by an excessive filtration due to the use of MMC.

The frequency of cataract operation was higher in the MMC group (35%) than in the no-MMC group (19%) (chi-square, $p = 0.03$). The results are summarized in Table 3.

**Discussion**

This study did not show any influence on IOP reduction when MMC was used during primary trabeculectomy. The use of MMC as an adjuvant in eye surgery was extensively published at the beginning of the 1990s. MMC has also been widely used in pterygium surgery. Theoretically, MMC reduced scarring at the ostomy in trabeculectomy, yielding better results after surgery. The Cochrane Database published a systematic review in 2005 providing support for the use of MMC in trabeculectomy. The review considered different indications and concluded that MMC could yield better results in trabeculectomies. Unfortunately for primary trabeculectomies, the review was based on just four studies with a reduced number of patients included and short follow-up. In 1997, Martini et al. published a small randomized clinical trial (RCT) including 60 eyes that were randomized to MMC 0.1 mg/mL or no-MMC. They found a significant difference in IOP after 1-year follow-up. However, in an RCT (published in 2006) conducted by Girma et al., they did not find any differences in IOP reduction after addition of MMC. Several fall series describing the results after addition of MMC were published, but these studies had short follow-up, small numbers of patients included and no control groups. Although MMC has been used for over 20 years, very few RCTs were found in the literature to support the use of MMC in primary trabeculectomies.

The general results (with or without MMC) after trabeculectomy from this study showed an average complete success of 67.5% (without eye drops) and an average qualified success of 77.5% (with and without eye drops). Trabeculectomy seems to be an excellent way to reduce IOP in glaucoma patients studied 2 years after surgery. The criteria for defining success (IOP $\leq 18$ mmHg or IOP reduction $\geq 30$%) were based on the recommendations of the WGA for moderate glaucoma. Both criteria are quite strict compared to the criteria used in clinical decisions. Considering that around 5.5% of patients were classified as a total failure due to the necessity of a new surgery, the clinical results of trabeculectomy were as high as 94.5%. There is a difference of 17% between the success criteria chosen for the study and clinical practice. Patients who did not reach levels of success according to the criteria selected were nevertheless classified as success in clinical practice.

Endophthalmitis was a common complication after trabeculectomy. In general, it was a transient problem, but in three cases in the MMC group, it lasted more than 3 months and resulted in hypotony maculopathy with poor visual outcomes. Hypotony maculopathy after using MMC was found.

### Table 2. Comparisons between MMC and no MMC groups regarding complete and qualified success.

|                                | Group 1 (no MMC), % | Group 2 (MMC), % | Test          | p   |
|--------------------------------|---------------------|------------------|---------------|-----|
| Complete success (without eye drops) criterion 1 ($\leq 18$ mmHg IOP) | 66.2                | 62.8             | Log-rank (Mantel–Cox) | 0.88 |
| Complete success (without eye drops) criterion 2 ($\geq 30$% IOP reduction) | 76.6                | 64.2             | Log-rank (Mantel–Cox) | 0.21 |
| Qualified success (with and without eye drops) criterion 1 ($\leq 18$ mmHg IOP) | 71.4                | 74.4             | Log-rank (Mantel–Cox) | 0.84 |
| Qualified success (with and without eye drops) criterion 2 ($\geq 30$% IOP reduction) | 80.0                | 84.4             | Log-rank (Mantel–Cox) | 0.60 |

MMC: mitomycin-C; IOP: intraocular pressure.

### Table 3. Complication rate after trabeculectomy.

|                                | No MMC N (%) | MMC N (%) | Test          | p   |
|--------------------------------|--------------|-----------|---------------|-----|
| Hyphema                        | 5 (6)        | 7 (8.3)   | Chi-square    | 0.41|
| Hypotony $\leq 5$ mmHg         | 20 (24.1)    | 39 (46.4) | Chi-square    | 0.0002a|
| Choroidal detachment           | 10 (12)      | 23 (27.8) | Chi-square    | 0.0007a|
| Cataract operation             | 16 (19)      | 30 (35)   | Chi-square    | 0.003a|
| Needling                       | 21 (25)      | 22 (26)   | Chi-square    | 0.82|

MMC: mitomycin-C.

*Comparisons were shown to be significant.
to be 8.9% in a 5 years’ retrospective study. No patients developed hypotony maculopathy in the no-MMC group.

Cataract formation after trabeculectomy is a well-known complication. This study showed a significant difference between the no-MMC (19%) and MMC (35%) group. These results are consistent with previous studies. Due to this study’s retrospective nature, it cannot be determined whether the increased number of cataracts had a direct relationship with the use of MMC. It is possible that more patients with cataracts before trabeculectomy were included in the MMC group. However, it seems reasonable that MMC could have some toxicity over the lens, inducing cataract formation.

The number of medications after surgery was reduced significantly. A total of 70% of patients were without any treatment after surgery. The rest of the patients (30%) still needed medication. The average number of medications among them declined from 3.65 ± 0.52 to 1.92 ± 0.74. Fewer medications translates to reduced adverse effects, costs and so on. The difference between the number of medications used in the no-MMC group (1.68 ± 0.67) and MMC group (2.16 ± 0.82) was significant (t-test; p = 0.04). These results can be attributed to the study design, which was clinically based.

The limitations of the study include its retrospective design, multiple surgeons and a significant group of patients suffering from exfoliation glaucoma, probably unrepresentative of the general population of patients with glaucoma. Therefore, the study results may not apply to patients suffering from primary open-angle glaucoma. Furthermore, patients were operated on during two different periods of time, adding a certain bias to the results, probably due to differences in types of medications. Another limitation of the study is that the time of the day for IOP measurements was not included in the medical records. The bleb appearance was recorded in a very few cases so no analysis could be done. Reibaldi et al. conducted a retrospective study and found reduced IOP in the group treated with MMC compared with a group treated with a balanced salt solution (BSS). In this study, no placebo group was included, adding a certain bias. Negative results usually raise the question of sample size. This study included around 80 patients in each group. Previous studies included around 60 patients in each group. The number of patients included in the study cannot explain the negative results. Furthermore, this study included four different criterion that were analysed, all showing negative results. The use of Kaplan–Meier curves increased the reliability of the study.

The dose of MMC used in this study (0.4 mg/mL for 3 min) was based on previous evidence and was the common dose that the general part of ocular surgeons still use in Sweden. Probably, the dose is too high for trabeculectomies in virgin eyes (untouched conjunctiva) and in patients with White ancestry. Unfortunately, the evidence in the literature is limited regarding the concentration and the time MMC should be used. No studies based on Scandinavian were found.

Previous studies included mostly POAG patients, and it is possible that the concentration of MMC used in this study was not high enough to make any difference in patients suffering from pseudoexfoliation glaucoma. However, the Swedish population is known for its tendency to have a low rate of tenoconjunctival and scleral fibrosis after filtering procedures. Comparisons between studies are difficult to perform due to different populations, techniques, endpoint definitions and so on.

Conclusion

In conclusion, this study showed no difference in IOP reduction when MMC was added to primary trabeculectomies in Swedish patients. An increased number of complications were detected in the MMC group.

Clinical significance

Although this is a retrospective study with its bias, results should indicate a possible absence of effect of MMC in primary trabeculectomies in Swedish patients. Further randomized control trials, including a more substantial number of glaucoma patients, are still needed to prove the beneficial effects of MMC in glaucoma surgery.

Declaration of conflicting interests

The author(s) declared no potential conflicts of interest with respect to the research, authorship, and/or publication of this article.

Ethical approval

Ethical approval for this study was obtained from The Gothenburg Ethical Committee (no. 1050-13).

Funding

The author(s) disclosed receipt of the following financial support for the research, authorship, and/or publication of this article: This work was supported by the Skaraborg Institute Foundation.

Informed consent

Written informed consent was obtained from all subjects before the study.

ORCID iD

Marcelo Ayala https://orcid.org/0000-0002-2556-7578

References

1. Ryskulova A, Turczyn K, Makuc DM, et al. Self-reported age-related eye diseases and visual impairment in the United States: results of the 2002 national health interview survey. Am J Public Health 2008; 98(3): 454–461.
2. Quigley HA and Broman AT. The number of people with glaucoma worldwide in 2010 and 2020. Br J Ophthalmol 2006; 90(3): 262–267.
3. Resnikoff S, Pascolini D, Etya’ale D, et al. Global data on visual impairment in the year 2002. *Bull World Health Organ* 2004; 82(11): 844–851.

4. Leske MC, Heijl A, Hussein M, et al. Factors for glaucoma progression and the effect of treatment: the early manifest glaucoma trial. *Arch Ophthalmol* 2003; 121(1): 48–56.

5. Burr J, Azuara-Blanco A, Avenell A, et al. Medical versus surgical interventions for open angle glaucoma. *Cochrane Database Syst Rev* 2012; 9: CD004399.

6. Kirwan JF, Lockwood AJ, Shah P, et al. Trabeculectomy in the 21st century: a multicenter analysis. *Ophthalmology* 2013; 120(12): 2532–2539.

7. Gedde SJ, Schiffman JC, Feuer WJ, et al. Treatment outcomes in the tube versus trabeculectomy (TVT) study after five years of follow-up. *Am J Ophthalmol* 2012; 153(5): 789–803.

8. World Glaucoma Association (WGA). *Guidelines on design and reporting of glaucoma surgical trials (WGA)* (eds T Shaarawy, M Sherwood and F Grehn). Amsterdam: Kugler Publications, 2009.

9. Palmer SS. Mitomycin as adjunct chemotherapy with trabeculectomy. *Ophthalmology* 1991; 98(3): 317–321.

10. dos Santos Martins TG, de Azevdo Costa ALF, Alves MR, et al. Mitomycin C in pterygium treatment. *Int J Ophthalmol* 2016; 9(3): 465–468.

11. Wilkins M, Indar A and Wormald R. Intra-operative mitomycin C for glaucoma surgery. *Cochrane Database Syst Rev* 2005; 4: CD002897.

12. Martini E, Laffi GL, Sprovieri C, et al. Low-dosage mitomycin C as an adjunct to trabeculectomy: a prospective controlled study. *Eur J Ophthalmol* 1997; 7(1): 40–48.

13. Robin AL, Ramakrishnan R, Krishnadas R, et al. A long-term dose-response study of mitomycin in glaucoma filtration surgery. *Arch Ophthalmol* 1997; 115(8): 969–974.

14. Szymanski A, Gierek-Lapinska A, Kozia M, et al. A fluorophotometric study of corneal endothelium after trabeculectomy using different concentrations of Mitomycin-C. *Int Ophthalmol* 1996; 20(1–3): 95–99.

15. Matsuda T, Tanihara H, Hangai M, et al. Surgical results and complications of trabeculectomy with intraoperative application of mitomycin C. *Jpn J Ophthalmol* 1996; 40(4): 526–532.

16. Girma T, Courtright P, Mengistu F, et al. A placebo controlled double blind clinical trial of mitomycin C in primary trabeculectomy in Ethiopian patients. *Ethiop Med J* 2006; 44(3): 253–256.

17. Kupin TH, Juzych MS, Shin DH, et al. Adjunctive mitomycin C in primary trabeculectomy in phakic eyes. *Am J Ophthalmol* 1995; 119(1): 30–39.

18. Singh J, O’Brien C and Chawla HB. Success rate and complications of intraoperative 0.2 mg/ml mitomycin C in trabeculectomy surgery. *Eye* 1995; 9: 460–466.

19. Neelakantan A, Rao BS, Vijaya L, et al. Effect of the concentration and duration of application of mitomycin C in trabeculectomy. *Ophthalmic Surg* 1994; 25(9): 612–615.

20. Zahid S, Musch DC, Nizioł LM, et al. Risk of endophthalmitis and other long-term complications of trabeculectomy in the Collaborative Initial Glaucoma Treatment Study (CIGTS). *Am J Ophthalmol* 2013; 155(4): 674–680.

21. Greenfield DS, Suner IJ, Miller MP, et al. Endophthalmitis after filtering surgery with mitomycin. *Arch Ophthalmol* 1996; 114(8): 943–949.

22. Bindlish R, Condon GP, Schlosser JD, et al. Efficacy and safety of mitomycin-C in primary trabeculectomy: five-year follow-up. *Ophthalmology* 2002; 109(7): 1336–1341.

23. Reibaldi A, Uva MG and Longo A. Nine-year follow-up of trabeculectomy with or without low-dosage mitomycin-c in primary open-angle glaucoma. *Br J Ophthalmol* 2008; 92(12): 1666–1670.

24. Lusthaus JA, Kubay O, Karim R, et al. Primary trabeculectomy with mitomycin C: safety and efficacy at 2 years. *Clin Exp Ophthalmol* 2010; 38(9): 831–838.

25. Perkins TW, Gangnon R, Ladd W, et al. Trabeculectomy with mitomycin C: intermediate-term results. *J Glaucoma* 1998; 7(4): 230–236.