Study of Different Methods and Concentrations of Mitomycin C Application in Subscleral Trabeculectomy in Rabbits

Thesis
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By
Karim Magdi Naguib
M.B.B.Ch., MSc. Ophthalmology

Supervised by

Prof. Dr. Mohamed Adel Abdel Shafik
Professor of Ophthalmology
Faculty of Medicine, Ain Shams University

Prof. Dr. Sanaa Abdel-Maged Sammour
Professor of Pathology
Faculty of Medicine, Ain Shams University

Prof. Dr. Ali Hassan Saad
Professor of Ophthalmology
Faculty of Medicine, Ain Shams University

Prof. Dr. Hany Mohamed Hassan El Ibiary
Professor of Ophthalmology
Faculty of Medicine, Ain Shams University

Faculty of Medicine
Ain Shams University
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تحت اشراف

الاستاذ الدكتور/ محمد عادل عبدالشفيق
استاذ طب و جراحة العيون
كلية الطب جامعة عين شمس

الاستاذ الدكتور/ سناء عبدالماجد سمور
استاذ الباثولوجى
كلية الطب جامعة عين شمس

الاستاذ الدكتور/على حسن سعد
استاذ طب و جراحة العيون
كلية الطب جامعة عين شمس

الاستاذ الدكتور/ هانى محمد حسن الابيارى
استاذ طب و جراحة العيون
كلية الطب جامعة عين شمس

كلية الطب
جامعة عين شمس
القاهرة
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LIST OF ABBREVIATIONS

- 5 – FU : 5 fluorouracil
- 5- CU : 5- Chlorouracil
- 5- FC : 5-Fluoro cytosine
- AC : Anterior Chamber
- AGIS : Advanced glaucoma intervention study
- ANOVA : Analysis of variance
- BAK : Benzalkonium chloride
- BSS : Balanced salt solution
- CAT : Cambridge antibody technology
- DNA : Deoxyribonucleic acid
- ECM : Extra cellular matrix
- H&E : Hematoxylin and Eosin
- HLA : Human lymphocyte antigen
- HTF : Human tenon fibroblasts
- IFN : Interferon
- IOP : Intraocular pressure
- M-6-P : mannose-6-phosphate
- MMC : Mitomycin C
- MMP : Matrix MetalloProteinase
- mRNA : messenger ribonucleic acid
- NIH : National Institutes of Health
- NTG : Normal Tension Glaucoma
- OGN :Oligonucleotide.
- P : Probability.
- PAS : Periodic Acid Schiff
- PBS : Phosphate-buffered saline
- PDT : PhotoDynamic Therapy
- POAG : Primary Open Angle glaucoma
- POE : Poly Ortho Esters
- SD : Standard deviation
- TGF-β : Transforming growth factor beta
- tPA2 : Tissue plasminogen activator
- tTgase : Tissue transglutaminase

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INTRODUCTION

Glaucoma is a leading cause of blindness worldwide and is the second most frequent cause of legal blindness in industrialized countries. In glaucoma the optic nerve is progressively damaged causing defects in the visual field, usually asymptomatic until the central vision is affected.

The goal of glaucoma management is to preserve the patient's quality of life. The only treatment option proved to prevent the loss of vision is to lower the intraocular pressure to a level deemed safe for the eye. Classically speaking, the recommended steps for lowering the intraocular pressure in primary open angle glaucoma (POAG) are topical medications first, followed by laser trabeculoplasty and, lastly, incisional surgery **(Traverso et al., 2005)**.

Surgical failures of trabeculectomies are in most cases caused by a wound healing response at the level of the episclera and the deep fibrovascular layers of Tenon's capsule that evoke a permanent increase of outflow resistance or even complete occlusion. While this obstruction is less common in patients with uncomplicated primary open angle glaucoma (POAG), it is a frequent complication in patients undergoing

repeated procedures or with other forms of glaucoma. The trabeculectomy success rates for POAG range from 24% to 75 % **(Mietz et al., 1998)**.

Recent data from the NIH advanced glaucoma intervention study (AGIS) have shown that individuals with the lowest intraocular pressures (average 12.3 mm Hg) had virtually no overall glaucomatous progression over nearly a decade. The healing response after surgery is the main long term determinant of long term post operative intraocular pressure. Therefore, if we are able to control the healing response in all patients after glaucoma surgery, it offers us the tantalizing prospect of minimal or no disease progression in the vast majority of our glaucoma patients, even those with advanced disease **(Khaw et al., 2002)**.

However, current studies do reveal that none of the current operations for glaucoma are totally ideal yet, and further research, particularly on surgical methods and wound healing control, is needed so that optimal long term pressure control can be achieved for all our patients with a minimum of complications **(Khaw et al., 2003)**.

Rabbit models of glaucoma filtration surgery, although commonly used experimentally, represent a very aggressive scarring response compared with that in humans. Therefore,

virtually every agent shown to reduce scar tissue formation in the rabbit is effective in humans **(Cordeiro et al., 1999(a))**.

Because of the aggressive wound-healing response in rabbits with surgical failure mostly occurring within 1 week, this animal model is believed to be equivalent to high-risk eyes in humans. The use of antifibrotic drugs, such as 5-fluouracil and mitomycin C, prolongs bleb survival. However, the effect is not permanent and, depending on drug concentration, it will subside within 2 weeks **(Grisanti et al., 2005)**.

Mitomycin-C has a profound inhibitory effect on the scarring response after glaucoma filtration surgery. Estimates suggest that intraoperative mitomycin C is used in at least 50% of primary trabeculectomies in the United States and Japan. The benefits derived from the antiscarring activity have, however, been tempered by a possible increase in postoperative complications, including chronic hypotony, bleb leak, and endophthalmitis **(Crowston et al., 2002)**.

The advantages of single dose intra operative administration of MMC include convenience, ease of administration and proven efficacy. However it is difficult to titrate dosage to each patient **(El-Harazi et al., 1999)**.

El-Harazi et al., in 1999 reported the effectiveness of low dose post operative application of MMC in rabbits.

An early description of MMC topical use appeared in the Japanese literature. **Kunimoto and Mori** in 1963 used a concentration of 0.04% three times daily for 1 to 2 weeks after pterygium surgery, with no recurrences. A number of subsequent articles from both Asia and North America have confirmed their good results, with pterygium recurrence rates of 2% to 16% **(Waller and Adamis, 2002)**.

Modulation of wound healing

Wound healing:

The conjunctival wound healing response is a major determinant of outcome following glaucoma filtration and squint surgery **(Addicks et al., 1983)**.

Wound healing is a complex and dynamic process of restoring cellular structures and tissue layers. The human adult wound healing process can be divided into 3 distinct phases: the inflammatory phase, the proliferative phase, and the remodeling phase **(Mercandetti, 2005).**

The inflammatory phase is characterized by the influx of neutrophils and monocytes to the wound area followed by lymphocytes and macrophages, and the release of a variety of growth factors and other molecules. Re-epithelialisation and an increase in fibroblast activity and number occur in the proliferative phase, as also does angiogenesis and the laying down of granulation tissue. The remodeling phase consists of fibroblasts mediating the processes of wound contraction, extracellular matrix deposition, degradation and modification