بسم الله الرحمن الرحيم

Wound Modulation in Trabeculectomy

By

Adel Abdel-Razik Farag MD
Professor of ophthalmology
Zagazig University

introduction

- The main determinant of long-term IOP after glaucoma filtration surgery is the healing response.
- Although antimetabolites have revolutionized glaucoma surgery, the use of these agents is still associated with complications.
- Moreover, because surgery can fail despite the use of powerful antimetabolites, there is a need for better agents with improved risk-benefit ratio

STAGES OF WOUND HEALING

Stage I : acute inflammatory response:
in which a fibrin clot is formed and neutrophils predominate.

2 days after injury, inflammatory cells produce the cytokines that set up the later stages of wound followed by macrophage migration that do:

*** Clears out the inflammatory debris.**
*** Secrete growth factors, which activate fibroblasts.**

Stage II : Fibroblasts & granulation tissue formation
, including the formation of new blood vessels.

Angiogenesis
is primarily driven by vascular endothelial growth factor.

Granulation
composed of glycosaminoglycans and both type I and type III collagen.

In early scar formation, type III collagen predominates;
in a mature scar, type I collagen predominates.

Some fibroblasts differentiate to myofibroblasts with resultant wound contraction.

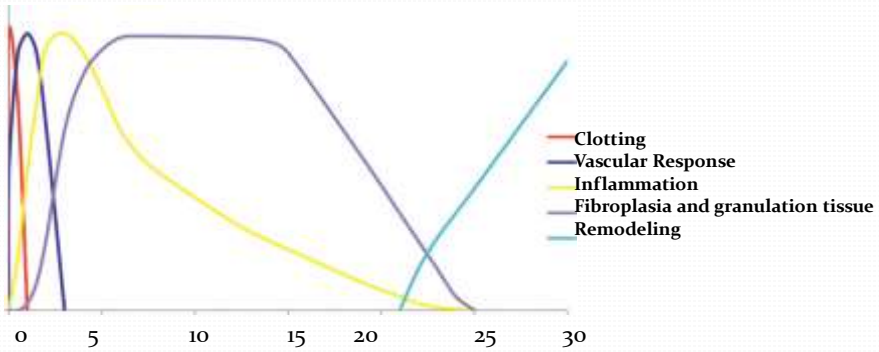
Stage III : wound remodeling

The matrix metalloproteinases remodel the fibrous and collagen components of the developing scar

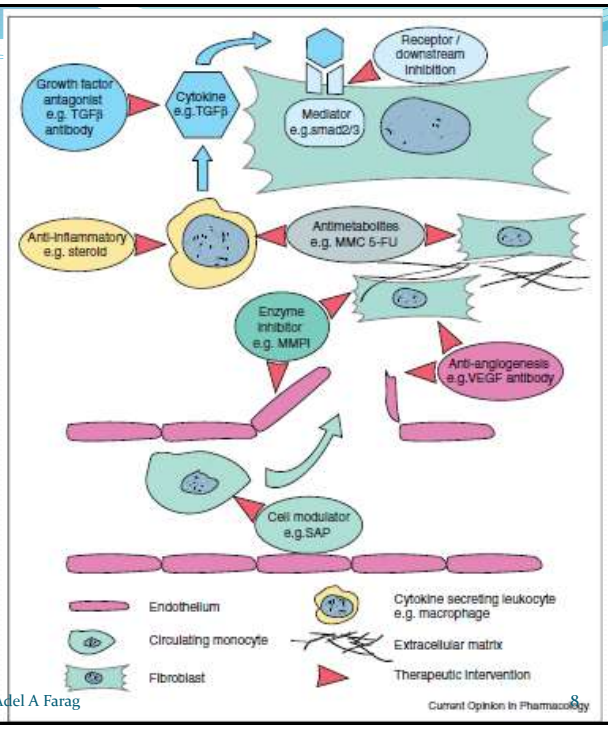
Characterized by:

* **Turnover of collagen in which new type I collagen is formed,**
* **Type III collagen is degraded, which increases the tensile strength of the wound.**
* **Constant turnover of collagen in the process of modulation till a mature wound is achieved .**

Overlap phases of wound healing (In relation to time)

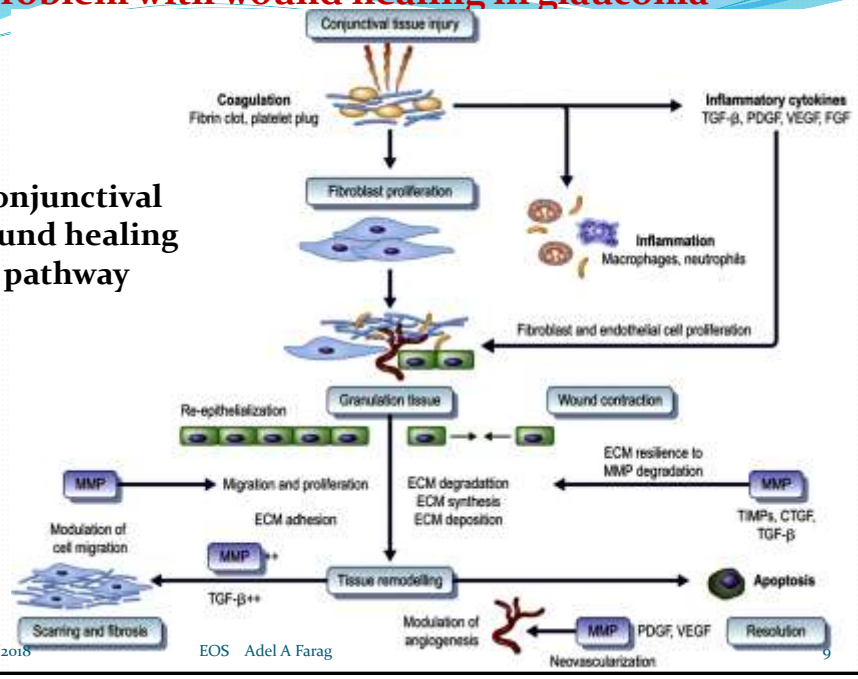


Pharmacological modulation of wound healing



Problem with wound healing in glaucoma

Conjunctival wound healing pathway



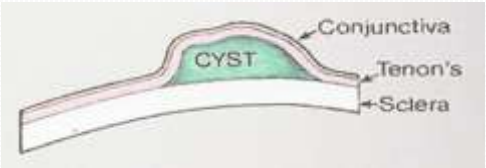
- Prognosis and success rates for glaucoma surgeries are largely dependent on wound healing and adhesion.
- Exaggerated wound healing, always results in the closure of the passage between the anterior chamber and the subconjunctival space .

- Tenon's fibroblasts are the main effector cells

involved in the initiation and mediation of wound healing and

fibrotic scar formation at the filtering bleb of trabeculectomy.

(Tenon's fibroblasts -→ myofibroblasts & TGF-b)



Methods of wound modulation

Traditional methods

Anti-inflammatory agents (steroid & NSAIDs)

Anti-fibrotic agents (MMC & 5FU)

New methods

***Anti-VEGF agents**

***CAT 152 (anti TGF-B)**

***Suramin**

*** Tranilast**

*** Signal pathway inhibition**

***Amyloid p**

***Anti-Sphingosine-1-phosphate (S1P) (Sonepcizumab)**

***Saratin**

***Si RNA gene silencing**

***Gene therapy**

***Radiation**

***Photodynamic therapy**

ANTI-INFLAMMATORY AGENTS

Steroids

Mechanism of action :

- Inhibit pro-inflammatory mediators (concentration, migration, and activity of leukocytes such as neutrophils and macrophages).

* Also reduce vascular permeability to minimize clot and fibrin production, and cellular migration to the site of injury

Methods of administration :

Topical and preoperative subconjunctival triamcinolone injections significantly improve the success rate of Trabeculectomy.

Non-steroidal anti-inflammatory (NSAIDs)

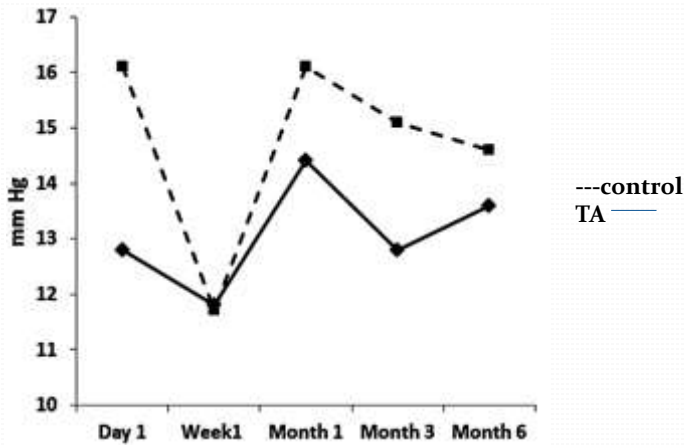
Mechanism of action

* Inhibition of the cyclooxygenase.
Thus prevents the conversion of arachidonic acid into the precursors of prostaglandins, thromboxane's, prostacyclin.
(pro-inflammatory compounds)

* Inhibits platelet function and clot formation

NSAIDs can used to avoid the known side effects of ocular steroid therapy such as cataract and infection.

Most of the studies found equivocal postoperative result, compared to steroid use after glaucoma filtration surgery.



IOP levels in postoperative follow up after Trabeculectomy with intracameral Triamcinolone Acetonide
Koval et al ; 2014

Antifibrotic

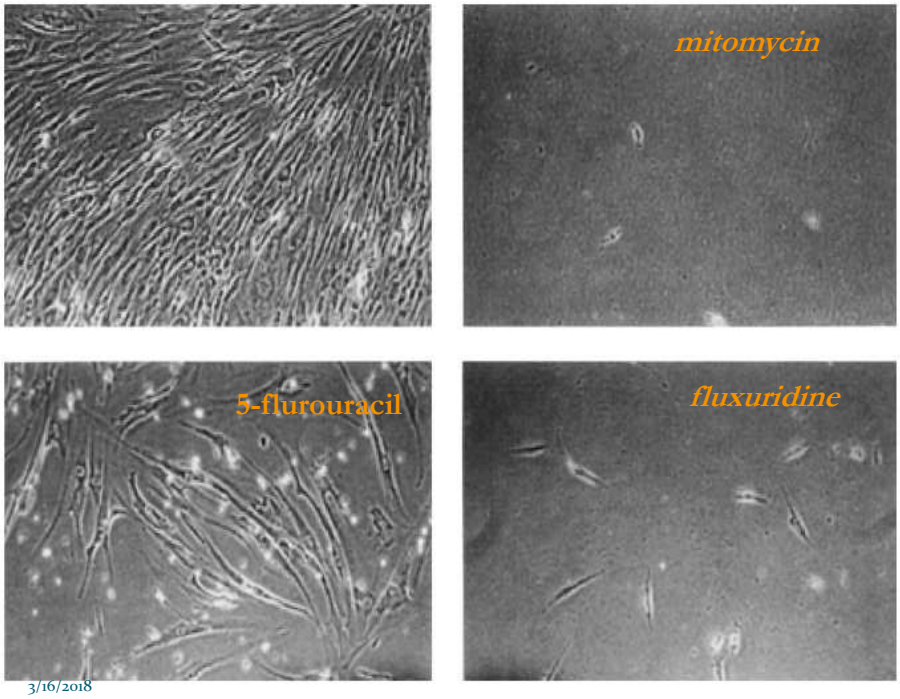
Prevent scar formation by alteration of fibroblast proliferation and activity.

- The use of ***antifibrotic agents*** to improve the success rates of trabeculectomy has become popular .
- The most commonly used agents are
  - **Mitomycin C (MMC)**
  - **5 Fluorouracil (5 FU)**

- These medications are used in conjunction with other preoperative, operative and postoperative medications to increase the success rate in glaucoma operations.

MMC

- Alkylating antitumor antibiotic
- MMC interrupt DNA replication → inhibits mitosis and protein synthesis
- It acts against all cells regardless its cycle.
- It has direct toxic effect on the overlying conj., ciliary epith, & inhibit angiogenic activity of BV
- Its potency is 100 times higher than 5 FU .



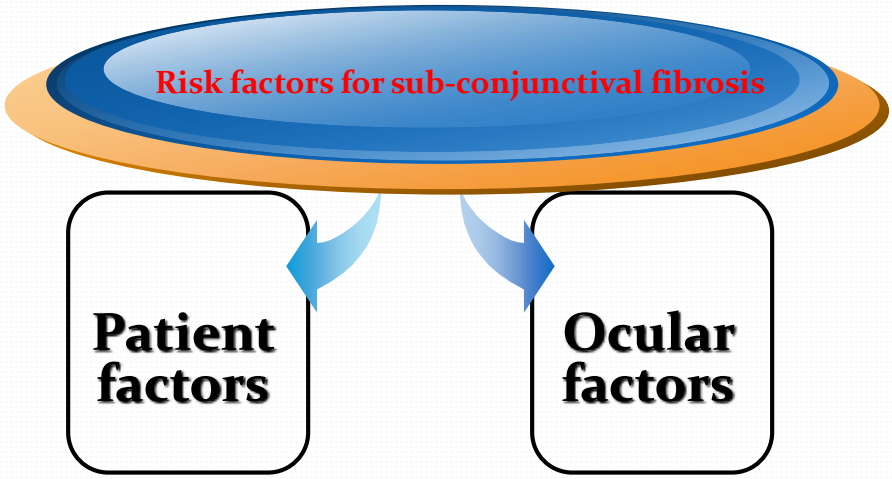
5 FU

- Flurinated pyrimidine analogue
- Inhibits thymidylate synthetaze → inhibit DNA synthesis .
- It acts selectively on the S phase of the cell cycle affecting the activity of replicating cells.

Complications Specific to use of 5 FU include:-

- Superficial punctate keratopathy
- Corneal & conj. Epith. Toxicity

Indications for anti-metabolites



Indications for antimetabolites

Patient factors

- ✓Young (< 40 years)
- ✓Black race
- ✓Previous chronic medical therapy *(especially with adrenaline)*
- ✓Previous failed trabeculectomy

Indications for antimetabolites

Ocular factors

- Secondary glaucomas
  (*neovascular and uveitic glaucoma*)
- Congenital /pediatric glaucomas
- Traumatic glaucomas
- Aphakic, pseudophakic glaucomas

Variables affecting the outcome

Fornix versus Limbus- Based Flap

- Both techniques can be used safely with antimetabolites
- fornix based flap may provide better long term outcome as cystic blebs with their related complications were found to be less common

RT eye a large area of MMC treatment. .diffuse, no cystic bleb was comfortable

5FU Vs Mitomycin C

- A single application of MMC appears to be superior to intraoperative use of 5FU especially in eyes at high risk glaucoma filtration surgery failure.

Conc. and duration of exposure

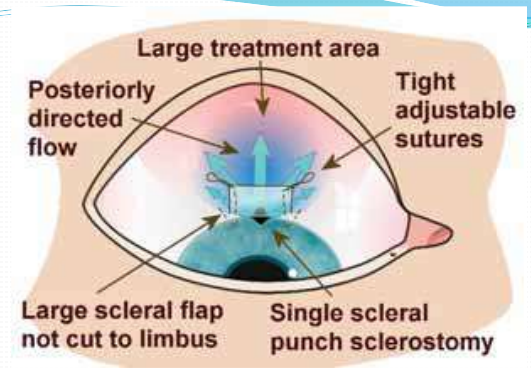
- Recommended conc. of MMC varies between 0.2- 0.5 mg/ml with variable outcomes
- Application of MMC 2-3 min. appears to have the same efficiency with 5 min. but application less than 2 min. Is suboptimal.
- Topical MMC 0.05mg/ml 4 times a day for 10 days post operative is an alternative technique of MMC application.

- A single intraoperative application of 5FU in a conc. of 50 mg/ml for 5 min. has prolonged localized inhibitory effects.
- Post operative 5FU injections can be given in addition to the intra-operative application.

  (5 mg daily. subconj. For 10 days , 180 degrees from the bleb

Sub scleral Vs Subconjunctival

- Greater IOP reduction was found in Trabeculectomy when adjunctive MMC was applied simultaneously under both scleral and conjunctival flaps.



Simple changes minimize the occurrence of complications after trabeculectomy , particularly when used in conjunction with strong antimetabolites such as MMC.

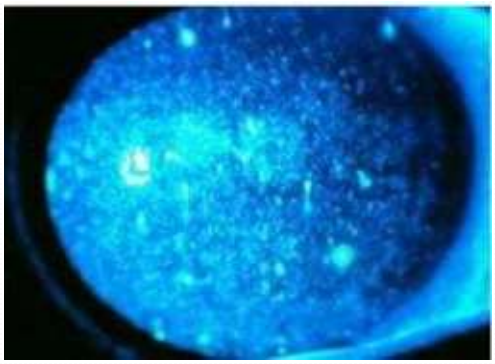
left eye.The bleb was small
and cystic, and he experienced discomfort
and recurrent

RT eye a large area of MMC treatment.
.diffuse, no cystic bleb was comfortable

Ring-of-steel
Anterior aqueous drainage occurs, and a ring of scar tissue forms

- In spite of these anti-metabolites increasing the success rate in glaucoma surgery, most ophthalmologists do' not use it in every glaucoma case because of the problems caused by these medications.

Complications of antifibrotics



Corneal epithelial defects (toxicity) especially with 5FU

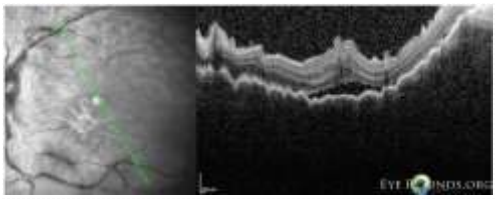
Cystic thin walled bleb especially with MMC that lead to :

Thin wall bleb can lead to chronic hypotony

Thin wall bleb can lead to bleb related late endophthalmitis rate increase to 2.1 % compared to 1% with non use

Other complications : include :

hypotony maculopathy is vision threatening complication associated with MMC use. Published rates vary significantly from 1.3-- 14%,



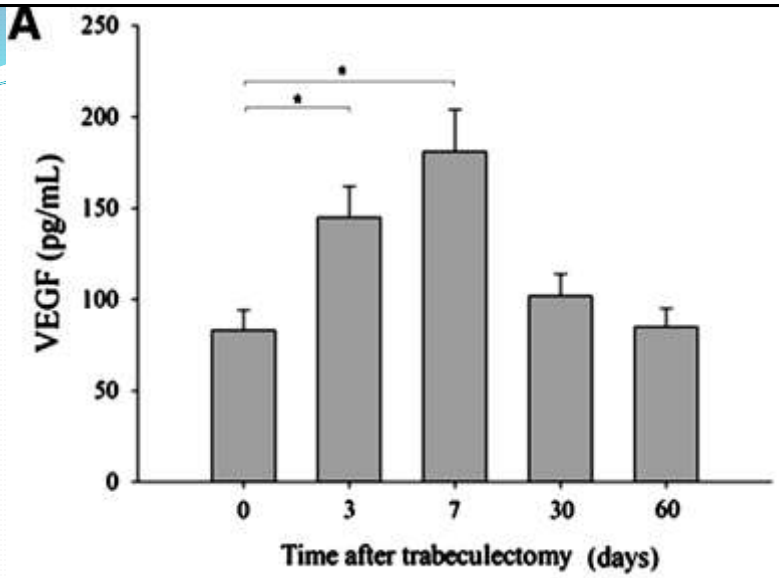
Other complications such as scleritis, scleromalacia, and endothelial cell loss reported with MMC

New Methods and Current Trends in Wound Modulation

Anti-VEGF Agens

Proliferative phase of wound healing, lead to angiogenesis that results in new blood vessels formation and the subsequent release of inflammatory mediators that lead to scar formation and bleb failure.

VEGF is present in large amounts after glaucoma filtering surgery and plays a key role in this process.



VEGF aqueous humor levels is higher in glaucomatous eyes and Significantly elevated after surgery

The use of anti-VEGF agents has expanded greatly over recent years to include neovascular glaucoma, diabetic macular edema, and corneal neovascularization

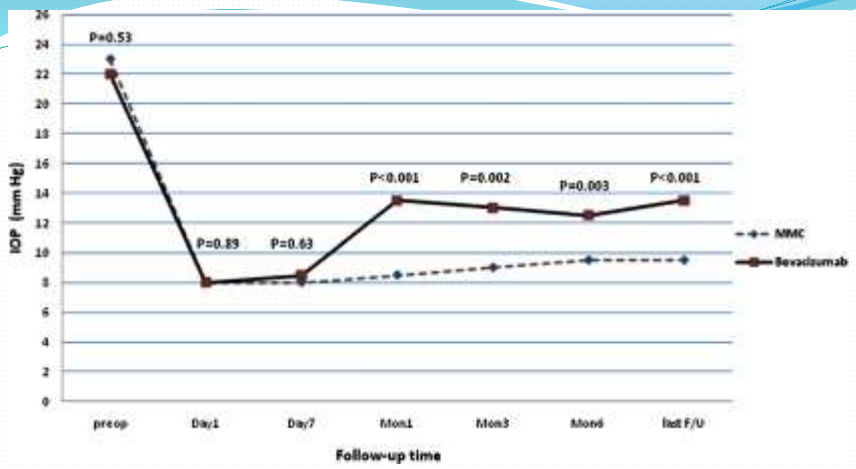
bevacizumab (Avastin)

is a monoclonal antibody against VEGF-A

Dose & Root of administration :

* Intravitreal (has more prolonged effect)
* Sub-conjuctivally

Dose : either 0.05 mg or 1.25 in 0.1 ml each
Success rate ranging from 90-95 %
which is comparable to MMC use



MMC versus bevacizumab in trabeculectomy of POAG
Nilforushan et al .2012

Ranibizumab (Lucentis)

Intraoperative Ranibizumab (0.5 mg) has the same results as MMC

But the bleb height and vascularity were significantly reduced in the Ranibizumab treated eyes,

A-MMC bleb

B- ranibizumab Bleb

CAT-152

- A novel monoclonal antibody (rh Anti -TGF-b2mAb) TGF is a potent fibroblast proliferation, collagen deposition and angiogenesis

Ocular disease processes like :

1-proliferative vitreoretinopathy,

2-cataractogenesis,

3-glaucomatous eyes and conjunctival scarring

have been associated with elevated TGF-b2.

Decorin :

Naturally occurring TGFb inhibitor, may be potentially safer because of the possibility of low immunogenicity

Suramin:

- A bispolysulfonated naphthylurea developed as an antineoplastic and anti parasitic agent.
- It inhibit a multitude of growth factors as PDGF, FGF, and TGF-b.136,
- *It is potent and can lead to chronic hypotony and choroidal detachment*

Tranilast

Anti-allergic agent, used in the treatment of allergic/atopic conjunctivitis and bronchial asthma through its inhibitory effects on the release of histamine.

It is found to inhibit other chemical mediators such as TGFb1, and PGE2 that inhibit fibroblast derived collagen synthesis

Topical dosing of 0.5 % Tranilast eye drops for 3 Months After filtering surgery as potent as MMC on human

Signal pathway inhibitors :

More recent work has been directed to block signal transduction pathway of which TGF-b exerts its inflammatory and proliferative effects in wound healing.

Various material as (SM-431542) still under experimental trial . It may provide a safer, more specific target of wound modulation in glaucoma surgery

AMYLOID P

Amyloid P (SAP) is a naturally occurring protein.
It is a potent modulator of the monocyte macrophage response ,
It inhibit pro-fibrotic cytokines and growth factors release

Experimental trial showed that it is superior than MMC in success of trabeculectomy and represent a promising agent in human

Anti-Sphingosine-1-Phosphate Monoclonal Antibody *(Sonepcizumab)*

S-1-P regulates a wide range of physiological processes including cell proliferation, migration, survival, and differentiation

Overproduction of S1P has been implicated in the processes of angiogenesis

(Sonepcizumab), is a humanized anti-S1P monoclonal antibody bind S1P results in a reduction of fibrotic activity

Still in experimental stage which is comparable to MMC

Saratin

Is a protein originally isolated from the saliva of the leech *Hirudo medicinalis*, but is now made by recombinant expression in yeast *(Hansenula polymorpha)*.

Its mechanism

1-Interfere with platelet integrin a2b1-collagen
2-Anti-von Willebrand factor-collagen binding,
3-preventing platelet aggregation and adhesion
5-its potent antithrombotic
6-Interfere with binding of inflammatory cells

******Still in experimental stage in glaucoma bleb trials***

siRNA GENE SILENCING

In glaucoma filtering surgery, siRNAs can be used to modulate the actions of Tenon fibroblasts and reduce scarring.

The siRNA molecule is then bound to cleaved RNA (by ribonuclease) leading to silence of RNA which is extremely precise mechanism of silencing unwanted genes.

The trial now are on human conjunctival and Tenon fibroblasts but in vitro with promising results

Gene therapy

Instead of blocking unwanted mRNA expression as siRNAs do, other methods of Gene therapy :

expressions of inhibitory genes on the cell cycle as a recombinant adenoviral vector for tumor suppression

p21, recombinant adenovirus vector for use in rabbit Trabeculectomy achieved target IOP levels for more time than MMC.

One consideration is the inherent risk of mutagenesis. Human trials have been undertaken thus far.

RADIATION

Single applications of b-radiation (750cGy) have significant anti- proliferative effect.

The radiation on human Tenon fibroblasts showed arrested growth in contrast to other antimetabolites, fibroblast migration and contraction were not inhibited,

The advantages

1-ease of application,

2-cost

3-Accessibility

Method :Strontium-90 applicator at the completion of surgery.

Disadvantages :

include the possibility of cataract formation and keratopathy.

PHOTODYNAMIC THERAPY

Offers selective modulation of wound healing in order to minimize side effects

Method :

injection of an inactive photosensitizing agent, the appropriate wavelength light is used to activate the compound, creating local oxidative tissue destruction.

The success rates

are high in experimental studies , no adverse effects were found. the use of PDT in glaucoma filtering surgery in human is still in its infancy.

Non drug wound modulation in glaucoma

Amniotic membrane transplantation :

**Amniotic membrane can promote epitheliazation of ocular surface

**it serve as a scaffold and a suitable basement membrane for epithelial cells to grow upon

act as an **inhibitor of fibrosis down regulating transforming growth factor-β signaling and myofibroblast differentiation.

Many studies found lower fibroblasts and macrophages cell counts in amniotic membrane assisted trabeculectomy

Conclusion

- In high risk of trabeculectomy failure → use antimetabolites.
- MMC is more potent and a single intraoperative application using 0.2 mg/ml for 3 min. is sufficient in most cases.
- Postoperative subconjunctival 5FU is helpful to promote survival of failing blebs.

- Histologic analysis showed that the multi-treatment blebs had more favorable characteristics when compared with MMC including
- less epithelial thinning, collagen thinning, and goblet cell loss.

- ***Numerous studies are needed to find the ideal combination drug regimen and with respect to human eyes response***

- *Surgeons' ability to fully modulate the healing process would ultimately allow them to improve the success rates and determine the long term final IOP in patients undergoing glaucoma filtration surgery.*

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Thank you
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