ENCAPSULATED FILTERING BLEB
A Selective Review—New Deductions

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SUMMARY

Filtering bleb encapsulation may, in some cases, be a severe complication following filtering surgery. The cause and mechanism of its development are not known. A selective review of data that might shed some light on these dilemmas, is presented. Based on these data, it is suggested that: (a) non-contractile collagen-producing fibroblasts play a major role in the process of bleb encapsulation, while in wound healing following filtering surgery, contractile fibroblasts are the major components; (b) the process of bleb encapsulation is less sensitive to the toxic effect of 5-Fluorouracil than wound healing; (c) collagen-producing fibroblasts may be less sensitive to the destructive effect of 5-Fluorouracil than contractile fibroblasts; (d) inflammatory mediators are important triggers of bleb encapsulation.

Encapsulated Bleb (EB) typically forms between the second and eighth week following filtering surgery. Its incidence has been reported following 8.3%-28% of these surgical procedures.[1-7] It consists of a fibrous tissue,[1] and presents as a tense, opalescent, thick-walled bleb, commonly associated with vascular engorgement of the overlying conjunctiva, and often accompanied by elevated intraocular pressure (IOP). If EB persists, and IOP remains high despite treatment by topical steroids, hypotensive medications and digital massage, surgical intervention is often indicated.[1-6] Although the mechanism and cause of BE is not known, some conclusions in this regard may be drawn from previous studies.

MECHANISM OF BLEB ENCAPSULATION

Wound healing versus bleb encapsulation

After surgical trauma, inflammation at the wound site occurs, followed by migration of fibroblasts which proliferate and produce collagen, elastin and mucopolysaccarides. It has been suggested that local tissue mesenchymal cells, or mesenchymal cells present in the blood, are the source of fibroblasts in this process.[8] Their attraction into the wound is aided by fibronectin and tissue hormones such as serotonin and prostaglandins. Fibronectin also assists in fibroblast adhesion to the damaged tissue.[9,10] There is evidence to support fibroblast heterogeneity with regard to proliferation and synthetic functions, including production of collagen.[11,12]

During wound healing, fibroblasts were detected in transparent ear chambers of half-lop rabbits in three separate zones: a proliferating zone, a 'synthetic' zone where they produced collagen, and a cross-linking zone where they were quiescent.[13] It has been further suggested that in wound healing, some fibroblasts are programmed specifically to synthesise collagen and others are also programmed to synthesise contractile protein, and become myofibroblasts.[14,15] This fibroblast-myofibroblast transformation is promoted by several factors such as tissue hypoxia, tension at the wound, and tension in the surrounding tissue.[16-18] The myofibroblasts have been identified as the major source of the contractile forces at the wound.[19] These cells have many features that are not shared by fibroblasts, but resemble those of smooth muscle cells.[20,21] However, it has been further demonstrated that myofibroblasts of normally-healed scars are different in character from those of pathologic conditions.[22]

The capsule of the EB behaves clinically as a relatively noncontractile tissue. It has been described as being pushed toward the conjunctiva by the aqueous pressure, creating a dome-shaped bleb.[3] This is different from bleb scarring, where flattening of the bleb wall is evident, due to myofibroblast contraction.[19] It is therefore suggested that non-contractile collagen-producing fibroblasts play the major role in the process of BE, while in wound healing following filtering surgery, contractile fibroblasts are the major components as well.

Bleb Encapsulation and 5-Fluorouracil

Analysis of the effects of 5-Fluorouracil (5-FU) in the development of both BE and filtering bleb scarring may be helpful in further evaluating the process of BE. 5-Fluorouracil is an inhibitor of fibroblast proliferation, and has

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also been demonstrated to cause intracytoplasmic vacuolar degeneration in existing, intravitreally-injected, dermal fibroblasts in rabbits,[23] and to cause morphologic changes in *in vitro* neonatal rat heart[24] and human Tenon's capsule fibroblasts.[25] It has been suggested that the mechanism of the toxic effect of 5-FU on (nonmalignant) fibroblasts operates through its incorporation into the RNA of the proliferating cells, whereas the inhibition of DNA synthesis, through the inhibition of thymidilate synthetase (as ocurs in cancerous cells), is minimal.[26]

Several reports have shown that treatment by repeated sub-conjunctival administrations of 5 mg 5-FU following filtering surgery did not inhibit post-operative EB, while it did inhibit bleb scarring (Table I). Ruderman and associates, in a randomised study, injected 5-FU following filtering surgery into 14 human eyes with various types of glaucoma with poor surgical prognosis; 12 control eyes did not receive 5-FU. Five mg 5-FU was administered once daily (or less) during the first post-operative week, and one injection during the second week.[7] This was considered (a "low dose" of 5-FU as compared to that administered in other studies.[27,28] Encapsulated bleb developed in 21.4% (3/14) of the 5-FU-treated eyes, but in only 8.3% (1/12) of the controls. Filtering bleb scarring, on the other hand, was far more common in the control eyes: 75% (9/12) versus 14.2% (2/14) in the 5-FU treatment group. Thus, a low dose of 5-FU in eyes prone to postoperative cicatrisation resulted in a higher incidence of BE than in that of the controls, while it inhibited bleb scarring. Wilson reported that 10 injections of 5 mg 5-FU, which were administered during three weeks following filtering surgery in eyes with poor surgical prognosis, had not reduced the incidence of BE, but had been successful in the inhibition of bleb scarring.[29]

In another randomised study, trabeculectomy was performed as an initial procedure in eyes with primary glaucoma. Four to six subconjunctival injections of 5 mg 5-FU administered into 25 eyes over 10 days, resulted in significant inhibition of bleb scarring, in comparison to 25 non-treated eyes. However, the rate of EB was 12% (3/25) in the 5-FU-treated group, and 8% (2/25) in the controls[30]. Thus, although 5-FU inhibited filtering bleb scarring, it did not inhibit BE in the three cited series.[7,29,30] We may deduce that the progress of BE is probably less sensitive to the inhibitory effect of 5-FU than scar formation.

These findings tend to support the assumption that different kinds of fibroblasts play the major role in the two pathogeneses. Examination under an electron microscope of a capsule of an EB which was partially excised from a bleb, seven weeks following filtering surgery with 5-FU, revealed many atypical fibroblasts. In spite of the morphologic changes, abundant amount of collagen was present in the matrix (Fig. 1) (unpublished data). These changes differ from those demonstrated in the inhibition of contraction band formation: 5-FU treatment following intravitreal injection of homologous rabbit dermal fibroblasts resulted in the inhibition of scar ('band') formation; intracytoplasmic vacuoles were detected in many fibroblasts, together with markedly reduced amount of collagen in the matrix.[23] One possible theory is that 5-FU is more toxic to contractile fibroblasts than to non-contractile collagen-producing fibroblasts; or, that the myofibroblasts in both pathologies differ in their characteristics as demonstrated in other tissues,[22] and thus in their reaction to different drugs. The first theory, however, may have support from another study:

5-Fluorouridine (5-FUR), the ribonucleotide metabolite of 5-FU, a more potent inhibitor of fibroblast proliferation and cell-mediated contraction than 5-FU, *in vitro* and *in vivo*, reduced the rate of intravitreal band formation in rabbits, but was substantially more efficacious in the inhibition of band contraction: In a controlled study, homologous rabbit dermal fibroblasts were injected intravitreally followed by 5-FUR injection. In the control non-treated eyes, intravitreal contraction band developed between the fibroblast injection site and the retina or optic nerve head (as previously described[31]) in 75% of the eyes. This band contracted, resulting in traction retinal detachment in 68.5% of eyes and in retinal pucker in 6.5% of eyes. In the 5-FUR-treated eyes, however, retinal detachment was not seen, while retinal pucker was noted in 29% of eyes.[32] Thus, in the eyes treated with 5-FUR, the rate of traction retinal detachment was significantly lower than in that of the controls, while the relatively minor contraction of the band caused only retinal pucker in part of these eyes. It is thus possible that (a) in the control eyes, the presence of an adequate amount of contractile fibroblasts in the contraction band resulted in strong retinal traction, i.e. tractional retinal detachment, whereas (b) in the 5-FUR-treated eyes, mainly contractile fibroblasts in the

Table I: Incidence of filtering bleb encapsulation following trabeculectomy with and without 5-FU.

Authors	Type of study	Type of eyes studied	Total 5-FU dose/time	Rate of bleb encapsulation	
				In the 5-FU treated eyes	In the control eyes
Ruderman & associates[7]	Randomised	Eyes with poor surgical prognosis	40mg/2w (or less)	21.4% (3/14)	8.3% (1/12)
Ophir & Ticho[30]	Randomised	Primary glaucomas	20-30mg/10d	12.0% (3/25)	8.0% (2/25)
Wilson[29]	Retrospective	Eyes with poor surgical prognosis	50mg/3w (or less)	'Inhibited bleb scarring but did not inhibit bleb encapsulation'	

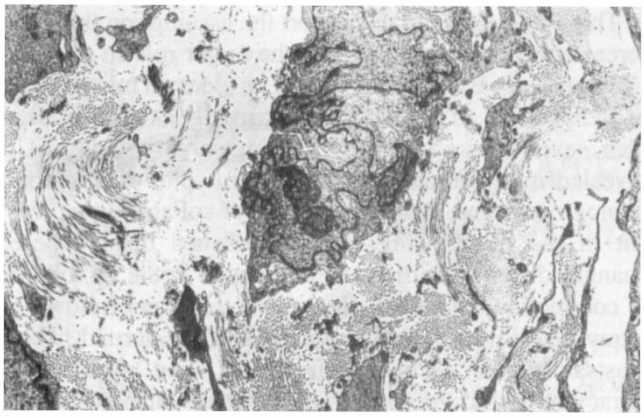

Fig. 1. *Examination of a partially excised capsule of a filtering bleb reveals atypic fibroblasts, with irregular cytoplasmic projections, markedly convoluted nuclei and large nucleoli. Abundant amount of collagen is seen in the matrix (EM × 5,500).*

band were affected by the drug. This could leave a higher proportion of collagen-producing cells in the band, resulting in a milder contraction, i.e. retinal pucker. It is thus similarly possible, as suggested above for 5-FU and filtering bleb, that 5-FUR, the potent metabolite of 5-FU, is more toxic to contractile fibroblasts than to those that only produce collagen. In the haematopoietic colonies, another nonmalignant proliferating system, it has been similarly suggested that variations in the intracellular metabolism of 5-FU as well as their proliferative rate probably determine differing sensitivities among the various types of colonies.[33]

INCIDENCE AND RISK FACTORS

The cause of BE is not known. It was found to develop more frequently in eyes that had previously undergone argon-laser trabeculoplasty (ALT)[4,5] or in eyes that had been treated for a prolonged time with a topical beta-adrenergic agent[5] or beta-adrenergic antagonist.[4] Other factors which were associated with increased risk of BE included male gender and history of prior BE.[5]

In eyes prone to post-operative filtering bleb scarring, such as those which have previouly undergone intra-ocular surgery, the incidence of BE is not clear. Van Buskirk reported on eight eyes with BE following filtering surgery: three eyes had recessed angles, two—pigmentary glaucoma, one—juvenile glaucoma, one—neovascular glaucoma and one—primary open-angle glaucoma. Seven of these eyes were considered to be prone to post-operative cicatrisation.[1] Richter and associates, however, reported a similar frequency of BE in eyes with previous ocular surgery as that in eyes with primary glaucoma (13.7%).[4] However, the incidence of bleb scarring, which might affect the clinical picture, was not reported. Since the incidence of bleb scarring is usually substantially higher in previously operated eyes than in primary glaucomatous eyes,[34] the possible 'masking' of encapsulation by a contractile scar that causes flattening of the bleb, makes the study on the incidence of BE in eyes with a higher tendency for bleb cicatrisation even more difficult. The possibility of 'masking' the encapsulation may be consistent with the observations of Ruderman and associates cited above (Table I), that more eyes with poor surgical prognosis developed BE following filtering surgery with low-dose 5-FU than without 5-FU treatment.[7] The incidence of BE was found to increase when surgery was performed in eyes with congenital or juvenile glaucoma.[4] It seems that relatively mild triggers of fibroblast activation, such as pre-operative ALT,[4,5] pre-operative prolonged use of various hypotensive medications,[4,5] previous ocular trauma[1] and surgery in congenital or juvenile glaucoma,[4] may promote BE following surgery; whereas stronger triggers such as previous intra-ocular surgery, promote bleb scarring.

Inflammation and Bleb Encapsulation

Mild anterior uveitis, which was concomitant with the development of BE, was detected 47 months and six months following filtering surgery in two patients. This rare occurrence suggests that inflammatory mediators triggered fibroblast proliferation and collagen formation and resulted in delayed BE. It is possible that such inflammatory mediators may similarly be an important factor for the more typical 'early' post-operative BE.[35] Lymphokines, secretory products of activated lymphocytes, influence *in vitro* fibroblast proliferation, migration and collagen synthesis.[36] *In vivo* depletion of T-lymphocytes by specific monoclonal antibodies results in impairment of that process.[37] However, depletion of the T-helper/effector lymphocyte subset has no effect on the process while depletion of the T-suppressor/cytotoxic subset actually enhances it.[38] It has been suggested that there is a subpopulation of T-lymphocytes (Thy–1.2$^+$, L3T4–, Lyt2$^-$) which normally stimulates fibroblast activation.[39,40]

Macrophages have also been shown to produce growth factor that can stimulate fibroblast chemotaxis and proliferation.[41] The monokines Interleukin-1 beta and tumour necrosis factor alpha can promote fibroblast proliferation *in vitro*.[41,42]

Other mediator molecules, such as platelet-derived growth factor, fibroblast growth factor, epidermal growth factor, and insulin-like growth factor-1 (IGF-1), have demonstrated mitogenic activity in fibroblasts,[43-46] but their physiologic relevance in *in vivo* models has not been elucidated. Insulin-like growth factor-1 (Somatomedin-C) which can also be synthesised by fibroblasts, has been identified in human wound fluid, and may act as a stimulus for fibroblast growth.[46] It is thus possible that some of the aforementioned inflammatory mediators take part in the process of fibrous tissue production and BE. The amount and/or characteristics of fibrous tissue that fibroblasts produce during BE, which is probably affected by cell number[47] as well by pre-operative factors,[4,5] and possibly by intra-operative and post-operative inflammatory factors, might determine the development and prognosis of EB.

Key words: Encapsulated bleb, Fibroblast, Fluorouracil, Glaucoma, Myofibroblast, Scar.

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