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The influence of scleral flap thickness, shape, and sutures on intraocular pressure (IOP) and aqueous humor flow direction in a trabeculectomy model

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Abstract

Purpose—IOP and aqueous humor flow direction determined by the scleral flap immediately after trabeculectomy are critical determinants of the surgical outcome. We used a large-scale model to objectively measure the influence of flap thickness and shape, and suture number and position on pressure difference across the flap and flow of fluid underneath it.

Methods—The model exploits the principle of dynamic and geometric similarity, so while dimensions were up to 30× greater than actual, the flow had similar properties. Scleral flaps were represented by transparent 0.8 and 1.6 mm thick silicone sheets on an acrylic plate. Dyed 98% glycerine, representing the aqueous humor was pumped between the sheet and plate, and the equilibrium pressure measured with a pressure transducer. Image analysis based on the principle of dye dilution was performed using MATLAB software.

Results—The pressure drop across the flap was larger with thinner flaps, due to reduced rigidity and resistance. Doubling the surface area of flaps and reducing the number of sutures from 5 to 3 or 2 also resulted in larger pressure drops. Flow direction was affected mainly by suture number and position, it was less towards the sutures and more towards the nearest free edge of the flap. Posterior flow of aqueous humor was promoted by placing sutures along the sides while leaving the posterior edge free.

Conclusion—We demonstrate a new physical model which shows how changes in scleral flap thickness and shape, and suture number and position affect pressure and flow in a trabeculectomy.

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Competing/ conflicts of interest

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Keywords

trabeculectomy; IOP; aqueous humor; sclera; model

Introduction

A trabeculectomy involves the creation of a guarded drainage channel in the sclera, through which aqueous humor can flow out of the eye into a subconjunctival bleb in a controlled manner, thereby reducing the intraocular pressure (IOP). Since its description in 1968^{1,2} there have been minor variations in the surgical technique, including different conjunctival incisions,^{3,4} different scleral flap shapes and sizes⁵⁻⁹ and different methods to create sclerostomies.^{5,9,10} From the square scleral flaps initially described, glaucoma surgeons now also use rectangular,^{4,9} semi-circular,⁵ and triangular⁶ flaps. A clinical study to compare a triangular flap in one eye versus a square-shaped one in the other eye of the same patient showed that there was no long-term difference in the post-operative IOP and complication rates.¹¹ However, this study was done before the higher incidence of cystic blebs and hypotony with the advent of antimetabolites.¹² Another study comparing triangular and square scleral flaps found that there was a pressure difference (lower for triangular flaps) for the first two post-operative days although this difference became non-significant after that. Additionally, in this study the triangular flaps only had one suture for 'high-flow' conditions, compared to four sutures for the square flaps for 'low-flow' conditions.¹³ Other studies have also considered the effect of varying the size of the scleral flap. In two laboratory studies modeling trabeculectomy on cadaver eyes with real time monitoring of the IOP, there was no significant effect of changing flap sizes on the eventual IOP.^{14,15} Two clinical studies showed no clear advantage or disadvantage of a larger scleral flap compared to a smaller one,^{7,16} although this was without the extensive use of antimetabolites. There are potential advantages of smaller flaps, there being a larger area of undisturbed tissue (which might be important in reducing overall bleb scarring or if repeat surgery is required) and a reduced astigmatic effect on the cornea,^{7,17} but there are also potential disadvantages such as poorer pressure regulation, a reduced diffusion area and also anterior placement of flow leading to more cystic blebs which would be more prominent with antimetabolite use.^{4,12}

A recent computational study modeled trabeculectomy surgery using finite element simulation.¹⁸ The authors compared the effect of various scleral flap and sclerostomy shapes and sizes on aqueous humor outflow rate in an objective manner. Computational analyses were performed based on fluid-solid interaction involving theoretical flap deformation caused by the flow of aqueous humor. The authors reported that larger scleral flap sizes and lower flap thicknesses led to increased aqueous humor outflow. Higher outflow rates were also associated with square flaps (compared to triangular ones), smaller sclerostomies (compared to larger ones) and semicircular sclerostomies (compared to circular ones). Finally, they also found that there was a linear relationship between pressure difference (between IOP and episcleral venous pressure [EVP]) and aqueous humor outflow rate.

Other strategies to improve the outcome of trabeculectomy also include the posterior diversion of aqueous humor flow. This appears to be associated with larger and more diffuse blebs with a lower incidence of cystic blebs, and a lower long-term risk of blebitis and endophthalmitis particularly when associated with antimetabolite use.^{4,9,19-21}

The purpose of this study was to use a large-scale mechanical engineering model to objectively measure the influence of flap thickness and shape, and suture number and position on both the pressure difference across a model scleral flap and the flow of fluid underneath it. It has been difficult to fully establish these principles using cadaver eyes because of the variability of tissue and surgical construction. The intended outcome was to develop a valid model to increase our understanding of how these different variables could affect the outcome of surgery. A model like this could potentially be used for refining surgical practice or for accelerating new adaptations. As far as we know, there has not previously been an approach such as this to understand the trabeculectomy procedure.

Methodology

Our physical model exploits the principle of dynamic and geometric similarity which is commonly used in the area of engineering to test fluid flow conditions with scaled models.²² Certain laws of similarity need to be observed in order to ensure that the model test data can be applied to the original. In addition to geometric similarity, dynamic (i.e. forces acting on the flow) similarity also need to be considered, where the velocity and streamlines of flow are matched. The dimensionless Reynolds number (Re) determines the ratio of viscous versus inertial forces. Dynamic similarity between a model and its original is realised when the Re for both are similar. The Re can be determined from

$$Re = ul/v,$$

where u is velocity, l is a characteristic length and v is kinematic viscosity. For aqueous humor flow from the eye, the Reynolds number,

$$Re \sim 10^{-3}.^{23}$$

In our model, with typical values for velocity, $u \sim 10^{-5}$ m/s, length, $l \sim 10^{-1}$ m and kinematic viscosity, $v \sim 10^{-3}$ m²/s,

$$Re \sim 10^{-3}.$$

As such, while the model is much larger than a scleral flap, the flow has similar properties. However, it should be noted that it is often impossible to achieve strict similarity during a model test.²⁴ The greater the departure from the actual conditions, the more difficult achieving similarity is. In these cases, some aspects of similarity may be neglected, focusing on only the most important parameters. In this experiment, some factors e.g. surface tension and angulation of flow have had to be ignored.

The main assembly consisted of 9 mm thick transparent acrylic upper and lower plates separated by an intermediate section (Figures 1a and 1b). The intermediate section was comprised of a back plate (to represent the limbus) and 1 cm × 1 cm × 1 cm cuboidal 'sutures' to adequately restrain the silicone sheets at their edges but allow space for their deformation (Figure 2a). The lower plate had a 10 mm inlet hole through which the working fluid flowed in underneath the sheet, injected using a syringe driver (Cole-Parmer, IL, USA). The function of the upper plate was just to apply uniform downward pressure on the intermediate section. This assembly was then mounted on top of a light box to visualize the flow. The images were recorded in black and white using a camera (Dolphin F145B; Allied Vision Technologies GmbH, Germany) connected to a personal computer; the camera was set to record images at pre-determined intervals of between 1 to 4 seconds after the start of flow. Experiments were carried out in a darkened room to enhance contrast for the image analysis data.

Silicone sheets (HT-6240; Rogers Corporation, CT, USA) of 0.79 and 1.60 mm (0.031 and 0.063 inch) thicknesses were used to model the sclera. These sheets had Young's modulus, $E = 1$ MPa (which is comparable to reported scleral values of 1.2-1.3 MPa²⁵) and Poisson's ratio, $\nu = 0.495$ (which is close to the 0.5 expected for an incompressible material). They were cut into 12 cm × 12 cm squares, 12 cm × 9 cm rectangles and 12 cm equilateral triangles. They were fixed at the 'limbus' end using the back plate of the intermediate section. Additionally, they were also fixed with 'sutures' in these configurations:

Rectangle - 2 sutures, 4 sutures, 5 sutures (Figure 2b).

Square - 2 sutures, 4 sutures, 5 sutures (same configuration as rectangle).

Triangle - 1 suture, 3 sutures (Figure 2c).

Before placement of each silicone sheet, a thin layer of glycerine was spread out on the bottom plate. The sheet was then placed and restrained with the back plate and 'sutures'. Next, the glycerine was squeezed out to create an air-tight seal between the sheet and the lower plate. The upper plate was then placed on top of the back plate and 'sutures', and secured with bolts and wing nuts. To prevent air from getting under the sheets again, the nuts were tightened to a torque of 2 Nm. The assembly was also checked to be level before the start of the experiment.

For the working fluid, we used 98% glycerine (VWR International Limited, UK) and undiluted Parker black ink (as a dye) in a ratio of 100:1. The dye was necessary for visualizing fluid flow underneath the flap. The kinematic viscosity of the fluid, ν was $\sim 1.15 \times 10^{-3} \text{ m}^2/\text{s}$ and thus suitable for use to maintain conditions of dynamic similarity. The solution was homogeneously mixed and the whole system ensured to be free of air bubbles before each run. A total of 60 ml of working fluid was pumped during each run; this volume was predetermined to be sufficient for equilibrium flow to develop.

Pressure measurement

To measure the pressure, we used a pressure transducer (model 26PCAFA6D, 15V excitation; Honeywell International, NJ, USA) connected to a signal conditioner/ amplifier (Mantracourt Electronics Ltd, Exeter, UK) and personal computer. Non-expansile tubing (outer diameter 6 mm, internal diameter 4 mm) was used throughout the system. This setup was connected to the inlet hole of the main assembly and syringe driver with a 3-way connector (Figure 1b). Once the complete setup was ready, the syringe driver was started and the pressure recorded, with the outcome of interest being the equilibrium pressure (Figure 1c).

Image processing and analysis

Images were processed using MATLAB. Prior to the experiment, calibration was performed by adding dyed glycerine into an empty beaker in increments of 0.5 ml up to a total of 20 ml, with serial images being taken (Figure 3a). With a known beaker diameter (59 mm), the average height of the fluid as the volume increased was calculated. MATLAB then measured the average intensity of each pixel in each image and correlated this with the average height of the solution. As the intensity increased with fluid height, a corresponding fluid height could be attributed to all areas in a black and white image (Figure 3b).²⁶ For the actual experiment images, MATLAB measured the average intensity (i.e. the fluid height) of each pixel in the initial (time = 0) and final (at equilibrium) images. It then subtracted the values of the initial image from that of the final image. The processed image showed the fluid height under the sheet in false-color (Figure 4).

A total of six runs was performed for each setting, and all combinations of the two thicknesses, three shapes, and three or two different numbers of sutures at flow rates of 100, 200 and 400 ml/h were tested. Statistical analysis was performed using Prism 4 software (GraphPad Software, Inc., CA, USA). The paired t-test was used when comparing two groups while repeated measures ANOVA with Bonferroni's Multiple Comparison post-test was used when comparing three groups. $P < 0.05$ was considered statistically significant.

Results

The pressures (in mmHg) under the silicone sheets give an indication of relative differences in IOP with different scleral flap configurations in the actual trabeculectomy procedure. Pressure drops are inversely related to these pressure values. We found that the pressure readings were heavily influenced by flow rate and sheet thickness. With our experimental setup, the lowest flow rate of 100 ml/h generally gave small pressure changes (< 0.1 mmHg) which were affected more by experimental errors. When a flow rate of 200 ml/h was used, this problem was reduced. The thinner 0.79 mm (0.031 inch) sheet also tended to provide low readings. The biggest differences were seen with the thicker sheet and rate of 400 ml/h. In Figures 5-7, error bars indicate 1 standard deviation (SD). Asterisks (*) indicate where differences were statistically significant ($p < 0.05$).

Influence of flap thickness

Pressures under the thicker sheets were higher than those under the thinner sheets, i.e. the pressure drops were smaller (Figures 5a and 5b) with the thicker sheets. When there were 3 sutures for triangles and 2 sutures for rectangles and squares, pressures under the thinner sheets were between 35.6-48.9% of pressures under the thicker sheets. When there were 3 sutures for triangles and 5 sutures for rectangles and squares, pressures under the thinner sheets were between 27.2-42.8% of pressures under the thicker sheets.

We found the fluid height under the thinner sheets to be higher for all three sheet shapes (Figure 5c). The difference was small with the rectangular and square sheets but larger with the triangular sheet. The direction of flow was similar between thicknesses.

Influence of flap shape

When there were 3 sutures for triangles and 2 sutures for rectangles and squares, we found that triangles maintained higher pressures, followed by rectangles and squares (Figure 6a). The pressures under the rectangular sheets were between 48.6-80.3% of pressures under the triangular sheets, and pressures under the square sheets were 29.7-40.2% of pressures under the triangular sheets. When there were 3 sutures for triangles and 5 sutures for rectangles and squares, a similar pattern was seen but the differences were all not significant (Figure 6b). Pressures under the rectangular sheets were between 61.8-108.9% of pressures under the triangular sheets, and pressures under the square sheets were 61.4-88.5% of pressures under the triangular sheets.

Our fluid height results were inconclusive when considering rectangles and squares with 5 sutures (see next section). With the 4-suture rectangle and square sheets, we found that the direction of flow was away from the limbus towards the opposite edge (Figures 6c i and ii). With the 2-suture rectangle sheet (Figure 6c iii), the direction of flow was away from the limbus and also to the sides. With the 2-suture square sheet (Figure 6c iv), the direction of flow was to the sides. With 3-suture triangles, the flow was also away from the limbus (Figure 6c v) but when there was only 1 suture then the flow was to the sides (Figure 6c vi).

Influence of suture numbers and positions

When the number of sutures was increased, the pressure drop became smaller (Figures 7a-7c). With the rectangular sheets, 4 sutures provided between 58.7-84.2% of the pressures with 5 sutures and 2 sutures provided between 44.6-130% of the pressures with 5 sutures. With the square sheets, 4 sutures provided between 45.3-98.4% of the pressures with 5 sutures and 2 sutures provided between 49.9-54.5% of the pressures with 5 sutures. With the triangular sheets, 1 suture provided between 52.5-81.0% of the pressures with 3 sutures.

With the 5-suture configurations, fluid tended to accumulate under the sheet as it was well constrained. With 4 sutures, the flow was directed more posteriorly, while with 2 sutures the flow was directed mainly to the sides (Figures 7d and 7e). With 3-suture triangles, the flow was away from the limbus (Figure 7f i) but when there was only 1 suture, then the flow was to the sides (Figure 7f ii).

Discussion

In the early post-trabeculectomy period, the scleral flap serves as the principal resistance to aqueous humor outflow.^{14,15,18} This makes it important as the key regulator of early intraocular pressure, determining the risk of hypotony. Conversely, it may also result in low reduction in IOP if it is poorly constructed. This study looked at pressure changes and aqueous humor flow direction afforded by this flap during this critical point after the procedure.

Influence of flap thickness

As scleral flap thickness increases, its rigidity and resistance to lifting also increases. As a result, with thicker flaps there is less aqueous humor flow below it and to the outside, and thus a smaller pressure drop occurs. Clinically, this relates to the thickness of the scleral flap which is created during trabeculectomy. In general, half thickness (around 250 μm thick) flaps are recommended.^{4,27} To facilitate more aqueous humor flow, thinner flaps can be created, and vice versa. However these scleral flaps cannot be made too thin as this predisposes to dehiscence or cheese-wiring of the flap and potentially low and uncontrolled IOP.^{4,9}

Influence of flap shape

With the 3 and 2 suture configuration comparison, the pressure differences were significant but this was not true with the 3 and 5 suture configuration comparison. This discrepancy could be due to the 2 suture square and rectangle flaps offering less resistance to flow compared to the 3 suture triangle flaps. However, in this same 3 and 2 suture configuration group, the differences between the rectangle and square flaps were also significant, so the effect of having less resistance in the 2 suture rectangle and square flaps cannot completely account for the significant difference. We thus conclude that triangular flaps of the same width and length result in lower pressure drops than rectangular or square ones. This is due to the smaller surface area of the triangular flap, being half of that of a rectangle or square with similar dimensions. The rectangles in our experiment also had lower pressure drops compared to squares, again due to the smaller surface area (108 cm^2 versus 144 cm^2). These findings support the results of Tse et al. which also showed that larger flaps create larger pressure drops.¹⁸ Clinically, this may be important in circumstances where a more gradual lowering of pressure is required e.g. myopic eyes which are more susceptible to hypotony or angle closure eyes where sudden lowering of pressure may predispose to aqueous misdirection. If larger reductions in IOP were needed, then the scleral flap would need to be made larger. However, there are practical limitations on infinitely increasing the widths of the scleral flaps. Too large a scleral flap may “use up” the upper sclera and conjunctiva, posing problems for further trabeculectomy or tube drainage implant placement, or may cause higher astigmatism.^{7,16}

We found that the direction of flow was influenced less by shape but more by the number and position of sutures, which will be discussed in the next section. With shapes, the small difference between them could be due to the similarity in their width:length aspect ratios, which in our experiment were 1.33 for rectangles, 1.00 for squares and 1.15 for triangles. In

a computational and experimental study by our group (unpublished data), we found that the width:length aspect ratio of a scleral flap was important in determining the direction of aqueous humor flow. With width:length aspect ratios of <2.0 , triangular flaps created more posterior flow. When this aspect ratio increased to >2.0 , the effect was reversed. For example, we found that changing the shape of the flap from a square (aspect ratio 1.0) to wide rectangle (aspect ratio 2.5) increases the fraction of fluid moving posteriorly by a factor of 6. Therefore the creation of a wider rectangular flap leads to much more posterior flow and may potentially help the creation of more posterior diffuse blebs.

Influence of suture numbers and positions

With a higher number of sutures, the pressure drop becomes smaller. With well-constrained flaps, there is less space and aqueous humor flow underneath it. Less aqueous humor escapes through the flap gap and thus a higher pressure is maintained. We were unable to determine flow direction with the 5-suture configuration as the fluid accumulated under the well-constrained sheets. With 4 sutures, the flow was directed more posteriorly while with 2 sutures the flow was directed more to the sides. In the triangle group, this pattern was also seen with 3 sutures and 1 suture respectively. The direction of flow naturally avoids the suture positions due to the higher resistance encountered. In the case of 2 sutures (for rectangles and squares) or 1 suture (for triangles), it is also affected by the distance to the closest edge of the sheet. A larger portion of fluid flows to the sides as they are closer to the sclerostomy as compared to the opposite edge.

Careful control of the pressure drop is important to avoid hypotony after trabeculectomy. However, if there is too little flow underneath the scleral flap, there might be increased scarring and resistance underneath it, which would lead back to poor aqueous humor outflow and high IOP. Clinically, one method that is being used to titrate this effect is with the use of adjustable or releasable sutures.^{28,29} The direction of the increased flow following adjustment can be determined by the positioning of the adjustable sutures posteriorly. Generally, a slightly higher post-operative IOP is desired as it is easier to lower pressure by adjusting or removing sutures after the initial surgery, compared to resuturing if the pressure is too low.

There were a number of limitations in our experiment. Suture tension can be an influential factor in determining pressure and direction of flow. We did not look into this effect and designed our experiment such that there was standardized and adequate but not tight suture tension at the edges of the scleral flap in our model. We also acknowledge that in the actual procedure, numerous other variables come into play, especially in the longer term e.g. wound healing and scarring under the conjunctival bleb.³⁰ As such, our study only provides information on the early post-operative control of IOP and aqueous humor flow.

Conclusion

Pressure changes and control of flow direction accorded by the scleral flap immediately after trabeculectomy are important to determine the surgical outcome. We demonstrate a large-scale physical model of a scleral flap, which is able to show how changes in flap thickness, shape and the number and position of sutures affect the pressure and flow characteristics.

With clinical judgment, changes in these variables can be made by surgeons to adjust for the intended outcome. Additionally, the model that we described may be used for refining surgical practice or for accelerating new adaptations.

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The principal investigator, Amir Samsudin confirms that he has full access to all the data in the study and takes responsibility for the integrity of the data and the accuracy of the data analysis.

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Figure 1.

a) Elevated view photograph of the main assembly with a triangular flap in position. b) Photograph of complete setup. c) Typical pressure curve showing equilibrium pressure. Small pulsations are seen due to the small pressure changes involved, and the relative insensitivity of the pressure transducer at low levels of pressure (<1 mmHg).

Figure 2.

a) Schematic diagram of silicone sheet from above and a cross section view. The sheet is indicated in grey and the back plate and 'sutures' in red. The inlet hole (sclerostomy) is indicated in blue and the bottom plate in white. W – width, L - length, T - sheet thickness, h - fluid height. b) Configurations for rectangles and squares - 2 sutures (left), 4 sutures (middle) and 5 sutures (right). c) Configurations for triangles - 1 suture (left) and 3 sutures (right).

a)

b)

Figure 3.

a) Calibration using dye increments. Dyed glycerine was added into an empty beaker, and serial images were taken. MATLAB determined the average intensity of each pixel in each image and correlated this with the average height of the fluid. As the intensity increased with fluid height, a corresponding fluid height could be attributed to all areas in a black and white image. b) Calibration curve for image processing. This indicated a linear relationship between the change in intensity (I_0/I) and the height of fluid (thickness) up to around 4 mm. The curve levelled out after that as the intensity became saturated and the program could not

register any further change. From this information, we set the maximum reading of the fluid height in the MATLAB script to 4 mm.

Figure 4. Method of processing by MATLAB. The initial image is subtracted from the final image and the intensity converted into fluid height in the processed image. Suture positions can just be visualized in the processed image.

Figure 5.

a) Comparison between thicknesses (3 sutures for triangles, 2 sutures for rectangles and squares). Error bars indicate 1 SD, while (*) indicate where differences are statistically significant. Pressures were higher with the thicker sheets. b) Comparison between thicknesses (3 sutures for triangles, 5 sutures for rectangles and squares). Pressures were higher with the thicker sheets. c) False color final images showing fluid height under (i) rectangle (ii) square and (iii) triangle sheets. Suture positions (grey boxes) are superimposed on the images. The images on the left are for 0.063 inch thickness while those on the right are for 0.031 inch thickness. Fluid heights under the thinner sheets were higher for all three sheet shapes.

Figure 6.

a) Comparison between shapes (3 sutures for triangles, 2 sutures for rectangles and squares). Error bars indicate 1 SD, while (*) indicate where differences are statistically significant. Triangles maintained higher pressures, followed by rectangles and squares. b) Comparison between shapes (3 sutures for triangles, 5 sutures for rectangles and squares). c) False color final images showing fluid movement under a (i) rectangle with 4 sutures, (ii) square with 4 sutures, (iii) rectangle with 2 sutures, (iv) square with 2 sutures, (v) triangle with 3 sutures and (vi) triangle with 1 suture. Suture positions (grey boxes) are superimposed on the images. With 4-suture rectangle and square sheets, the direction of flow was away from the limbus towards the opposite edge. With the 2-suture rectangle sheet the flow was away from the limbus and also to the sides. With the 2-suture square sheet, the direction of flow was to the sides. With 3-suture triangles, the flow was also away from the limbus but when there was only 1 suture then the flow was to the sides.

Figure 7.

a) Comparison between number of sutures in the rectangle group. Error bars indicate 1 SD, while (*) indicate where differences are statistically significant. Generally, higher pressures were maintained with more sutures. b) Comparison between number of sutures in the square group. Generally, higher pressures were maintained with more sutures. c) Comparison between number of sutures in the triangle group. Higher pressures were maintained with more sutures. d) False color final images showing fluid movement under a rectangular sheet with (i) 5, (ii) 4 and (iii) 2 sutures. Suture positions (grey boxes) are superimposed on the images. With 5 sutures, fluid accumulated under the sheet as it was well constrained. With 4 sutures, the flow was directed posteriorly while with 2 sutures the flow was directed posteriorly and to the sides. e) False color final images showing fluid movement under a square sheet with (i) 5, (ii) 4 and (iii) 2 sutures. With 5 sutures, fluid accumulated under the sheet as it was well constrained. With 4 sutures, the flow was directed posteriorly while with 2 sutures the flow was directed to the sides. f) False color final images showing fluid movement under a triangular sheet with (i) 3 sutures and (ii) 1 suture. With 3 sutures, the flow was away from the limbus but with 1 suture it was to the sides.