Comparison of anterior capsulotomy techniques: continuous curvilinear capsulorhexis, femtosecond laser-assisted capsulotomy and selective laser capsulotomy

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ABSTRACT

Purpose To compare the anterior capsulotomy edge tear strength created by manual continuous curvilinear capsulorhexis (CCC), femtosecond laser-assisted capsulotomy (FLACS), and selective laser capsulotomy (SLC).

Setting Singapore National Eye Centre, Singapore and Excel-Lens, Livermore, California, USA.

Design Three arm study in paired human eyes.

Methods Capsulotomies were performed in 60 cadaver eyes of 30 donors using CCC, Victor Femtosecond Laser, (Bausch & Lomb, Rochester, New York, USA) or CAPSULaser, (Excel-Lens, Los Gatos, California, USA). Three pairwise study groups each involved 10 pairs of eyes. Study group 1: SLC eyes compared with fellow eyes with CCC. Study group 2: CCC eyes compared with fellow eyes with FLACS. Study group 3: FLACS eyes compared with fellow eyes with SLC.

A shoe-tree method was used to apply load to the capsulotomy edge, and Instron tensile stress instrument measured distension and threshold load applied to initiate capsule fracture. Relative fracture strengths and distension of CCC, FLACS and SLC were determined. Scanning electron microscopy (SEM) of capsule edges were reviewed.

Results Anterior capsulotomies behave as non-linear elastic (elastomeric) systems when exposed to an external load. The pairwise study demonstrated that the SLC fracture strength was superior to that of CCC by a factor of 1.46-fold with SLC 277±37 mN versus CCC with 190±37 mN. Furthermore, CCC fracture strength was superior to that of FLACS by a factor of 1.28-fold with CCC 186±37 mN versus FLACS 145 ± 35 mN (p < 0.001). This was determined by statistical analysis utilising the Wilcoxon matched-pairs signed-ranks test and in accordance with the Consolidated Standards of Reporting Trials guidelines. The capsule edge of SLC on SEM demonstrated a rolled over edge anteriorly and an alteration of collagen.

Conclusions The strength of the capsulotomy edge for SLC was significantly stronger than that of CCC which and both were significantly stronger than FLACS. The relative strengths can be explained by SEM of each type of capsulotomy.

INTRODUCTION

Cataract surgery is the most commonly performed surgical procedure in the world with over 25 million surgeries performed globally. 1 Cataracts also account for the majority of curable world blindness 2 and with improved small incision techniques the surgery is becoming safer and more reliable. The burden of cataract surgery remains a problem mainly in long-income and middle-income countries however, with the ageing population in industrialised nations, is likely to increase considerably in the next two decades. Furthermore, with the advent of high performance multifocal and extended depth of focus lenses, lens extraction is becoming increasingly popular as a means of vision correction. Increasing volume, improvements in efficiency, reproducibility and reliability of cataract surgery with reduced complications are motivating factors for further innovation.

Modern cataract surgery is performed through a small incision either by phacoemulsification or whole lens removal through a capsular opening made by a continuous curvilinear capsulorhexis (CCC). 3 Over the last 10 years, the femtosecond laser in combination with anterior segment imaging has been used to create a consistently circular capsulotomy of a defined size with good centration. 4, 5 Additionally other devices have been developed including a radio frequency cautery through a disposable device containing a circular nitinol alloy ring placed in direct contact with the anterior capsule (Zepto Mynos Cellar Devices, Fremont, California, USA). 6, 7 A selective non-contact laser capsulotomy device has also been developed and produces a capsulotomy of any size in 1 s and works by laser energy absorption of the trypan blue dye stained anterior capsule (online supplementary video).

The ideal capsulotomy is one that can be performed rapidly and in a reproducible manner with good centration on the crystalline lens, circular with good edge strength whereby there is little or no risk of radial anterior capsular tears during cataract surgery, lens prolapse and manipulation. Radial anterior capsule tears can lead to intraocular lens instability and at worst could during cataract surgery extend posteriorly to involve the posterior capsule with subsequent vitreous loss in 50% of cases. 8, 9

Different methods of anterior capsulotomies have different resulting metrics including the accuracy of placement and centration as well as capsular edge strength. Capsular strength is a factor that can be influenced by a number of variables including...
method of capsulotomy used and capsulotomy size.\textsuperscript{10} We describe the comparative performance of three different techniques of capsulotomy specifically evaluating capsule strength and distension.

**MATERIALS AND METHODS**

Three preclinical pairwise studies of ex vivo human cadaver eyes were performed to compare relative threshold forces to initiate an anterior tear for capsulotomies formed by selective laser capsulotomy (SLC), CCC and femtosecond laser-assisted capsulotomy (FLACS) techniques. Specifically, the pairwise comparisons were divided into three study groups: Study group 1: SLC to CCC, Study group 2: CCC to FLACS and Study group 3: FLACS to SLC.

In all groups, the left and right cadaver eyes were randomly allocated to the respective arms of the study. Pairs of phakic cadaver eyes from a donor in the specified age range of 35–70 years old were obtained within 72 hours postmortem from either the SightLife Eye Bank (Seattle, Washington, USA) and Singapore Eye Bank (SNEC, Singapore).

In all eyes of all groups, to improve access and visibility as well as optical measurement of the capsulotomy, the cornea and iris was removed before performing the relevant procedure. In all eyes, a cohesive ocular viscoelastic device (OVD) (2% sodium hyaluronate acid, CAPSULVisc 2%, Excel-Lens, Los Gatos, California, USA) was used. In all eyes, the lens nucleus was following capsulotomy hydro-expressed using balanced salt solution (BSS) and resultant capsular bags filled with dispersive OVD (3% sodium hyaluronate acid, CAPSULVisc 3%, Excel-Lens, Los Gatos, California, USA). If cortex was observed it was removed before performing the relevant procedure. In all study eyes using image processing software (provided by Stellaris, Bausch & Lomb, Rochester, New York, USA).

**Selective laser capsulotomy**

The SLC technique involved initially rinsing the anterior capsule with BSS (Alcon, Fort Worth, Texas, USA). The cannula tip of the trypan blue syringe was placed centred on the anterior capsule and 0.15 mL of trypan blue ophthalmic surgical solution (CAPSULBlue, Excel-Lens, Los Gatos, California, USA) applied with a gentle pooling technique. This was left for 10 seconds and then rinsed with 5 mL of BSS to remove any unabsorbed dye. The cohesive 2% OVD was then applied to the anterior capsule filling from one side in a continuous manner to the other, similar to the back-fill technique used with OVDs in surgery. The patient interface lens was placed centred on the anterior capsule and OVD injected under the patient interface (CAPSULaser focusing aid) to remove any air bubbles. The CAPSULaser focusing aid ensured that the laser was focused at the plane of the anterior capsule. A projected laser reticule allowed the location of the desired capsulotomy to be centred on the limbus or as appropriately selected by the surgeon. The capsulotomy diameter was set to 5.0 mm in all cases. The laser footprint was depressed activating the laser to fire with one continuous pulse (rather than multiple pulses) for one second to create a 5.0 mm diameter capsulotomy. The patient interface was removed, and region of the capsulotomy was rinsed with BSS and the capsulotomy disc removed with forceps.

**Continuous curvilinear capsulorhexis**

With the CCC technique, the anterior capsule was similarly rinsed with BSS and OVD applied as described above. The CAPSULaser device was also utilised to project a red laser circular reticule of 5.0 mm on to the anterior capsule to facilitate a guide for the size, circularity and centration of the CCC. The CCC was formed in a continuous manner using forceps under OVD. Once completed the anterior capsule was rinsed with BSS and the capsulotomy disc removed with forceps.

**Femtosecond laser assisted capsulotomy (FLACS)**

The FLACS technique was performed by rinsing the anterior capsule with BSS followed by application of the OVD. A rigid contact lens and cohesive OVD were utilised to simulate the cornea and anterior chamber. The contact lens was centred on the anterior capsule and the laser patient interface placed on it with hydroxypropyl methylcellulose (CAPSULgel, Excel-Lens, Los Gatos, California, USA). The eye was then docked to the femtosecond laser (Victus, Bausch & Lomb, Rochester, New York, USA). The femtosecond laser was used at a pulse frequency of 80 kHz, pulse duration of 400–500 fs and near-infrared wavelength of 1040 nm. The laser energy setting was 7.0 μJ with spot and layer separations of 6 and 4 μm, respectively. The capsulotomy size was set to an intended diameter of 5.0 mm. Once the capsulotomy was accomplished, the patient interface was removed, and region of the capsulotomy was rinsed with BSS and the capsulotomy disc removed with forceps.

**Dimensional and stress evaluation analysis**

The capsulotomy perimeter and circularity were measured for all study eyes using imaging processing software (provided by Excel-Lens, Los Gatos, California, USA). A microscope mounted camera captured images of the capsulotomy with an intraocular ruler placed in the plane of the anterior capsule. The ruler was used to calibrate the dimensional scale. The capsulotomy rim edge was defined by the reviewer identifying at least 40 points. The least-squares method was used to determine best-fit-circle for these identification points: yielding the capsulotomy perimeter and centre.

The methodology for determining the load-extension curves was with an Instron 3343 mechanical tester (Instron Corp, Canton, Massachusetts, USA). The system was calibrated prior to use by the manufacturer. Measurements were performed using a standardised method as described in detail by Daya et al.\textsuperscript{10} In summary a uniquely designed wax-coated (to reduce probe friction on the capsular edge) probe which exerted maximal force and extension on the capsular rim only was inserted into the capsular bag through the capsulotomy with the rim located on the waist of the probe. The capsular bag supported by the probe was dissected from the globe with scissors cutting the zonules similar to the method described by Werner et al.\textsuperscript{11} The probes were then connected to the Instron 3343 mechanical tester for tensile strength testing. The system was reset to zero extension and the test commenced under computer control and video monitoring. One arm of the fixture remained fixed in position while the other was translated stretching the capsulotomy to its tear point. The capsular exterior of the capsular bag was frequently irrigated with BSS during the procedures to prevent dehydration. Both stress and magnitude of extension were recorded for each increment of the stepper motor. The rupture point for stress and extension were determined at the point of sudden drop in load. The threshold perimeter was calculated as twice the probe extension plus 4.5π for the curve probe perimeter.

Taking into consideration the influence of capsulotomy size and continuity on threshold load and the incidence rate of anterior tears\textsuperscript{10} the size and continuity was controlled in this study by excluding pairs of eyes based on dimensional analysis. A pair of
eyes was rejected from analysis if one or both eyes demonstrated one of the following three criteria:
1. Poor circularity as defined by non-circularity $4\pi$-area of the best-fit-circle/perimeter distance from point to point being less than 90%.
2. Poor circularity based on the method of minor and major axis ratio where the minor axis was less than 90% of the major axis.
3. Diameter sizing rejection criteria where the diameter was outside the range of the intended desired diameter of 5.0 mm with a tolerance of $\pm 0.2$ mm.

The datasets were analysed to test superiority between the two arms for each pairwise study utilising the Wilcoxon Signed Ranks Tests. The null hypothesis for each of the three pairwise studies was that the mean difference including CI between pairs was zero, with the alternative hypothesis being the mean averages were different to the extent one arm is proven to be superior and the other arm is proven to be inferior (two-sided).

### Scanning electron microscopy

The capsulotomy rim structures were evaluated for SLC, CCC and FLACS using scanning electron microscopy (SEM; Nanocraft, Los Gatos, California, USA). Anterior capsule tissue samples were collected for each capsulotomy technique. Some SLC samples were also purposefully cross-sectioned by freezing in liquid nitrogen and then sectioned with a stainless-steel histology blade to investigate the nature of the capsulotomy edge in more detail. The capsules were washed in Phosphate Buffered Saline for 60 min before being immersed in 1% aqueous solution, fixed in glutaraldehyde, then rinsed in distilled water, and introduced into osmium tetroxide for 4 hours at room temperature. Following this, the samples were dehydrated in increasing concentrations of ethanol. When the samples were critical point dried they were sputter coated with 8–10 nm thick layer of gold and examined using a scanning electron microscope (Vega 3, Tescan, Czech Republic).

### RESULTS

The donor ages and genders for the three pairwise studies are reported in table 1. All three studies had similar age and gender distributions with no meaningful differences.

The distributions for the capsulotomy extensions, perimeters and loads at the tear facture are reported in table 2, for the three pairwise studies. The mean load required to tear the anterior capsule in Study 1 was $190\pm37$ mN for CCC versus $277\pm38$ mN for SLC. The threshold perimeter for CCC at $25\pm2$ mm was also lower than SLC $29\pm2$ mm as was extension at $5\pm1$ mm for CCC versus $7\pm1$ for SLC. In terms of perimeter stretch, which is defined as the increase in perimeter relative to the original capsulotomy size, CCC had $59\%\pm20\%$ and SLC had $85\%\pm15\%$. This indicates greater distensibility of the anterior capsule following SLC compared with CCC (table 2). In Study group 2 where CCC was compared with FLACS, the tear load to cause a capsule tear was lower for FLACS ($145\pm35$ mN) than CCC ($186\pm37$ mN) and the perimeter and perimeter stretch were slightly less for FLACS ($23\pm2$ mm, $46\%\pm24\%$) than CCC ($24\pm2$ mm, $53\%\pm22\%$) while extension was similar in both groups at $5\pm1$ mm. In Study group 3 comparing FLACS to SLC the SLC treated capsules required a greater load ($269\pm38$ mN) than FLACS ($148\pm31$ mN), along with larger extension perimeter stretch (and perimeter) $78\%\pm16\%$ ($28\pm1$ mm) for SLC versus $46\%\pm24\%$ ($23\pm1$ mm) for FLACS. The load, perimeter and extension for CCC in Study 1 and 2 were similar as was FLACS in Study 2 and 3 and SLC in Study 1 and 3, demonstrating consistency of strength and distensibility between subgroups for each type of capsulotomy (table 2).

To facilitate the Signed Rank Tests, the pairs were organised in ascending absolute difference in perimeter stretch between the pairs for each of the three studies.

In Study group 1 where SLC was compared with CCC $90\%$ of the 10 pairs the perimeter stretch in SLC exceeded that of CCC, with pair number 1 being the exception. For the load parameter, all 10 pairs of SLCs exceeded that of the CCCs. Using Wilcoxon Signed Ranks Test statistical analysis results indicated SLC was

### Table 1  Donor age and gender distributions for each study group

<table>
<thead>
<tr>
<th>Study 1: SLC versus CCC</th>
<th>Study 2: CCC versus FLACS</th>
<th>Study 3: FLACS versus SLC</th>
</tr>
</thead>
<tbody>
<tr>
<td>Pair number</td>
<td>Age (years)</td>
<td>Gender</td>
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<tr>
<td>1</td>
<td>68</td>
<td>M</td>
</tr>
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<td>2</td>
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<td>F</td>
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<td>3</td>
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<td>F</td>
</tr>
<tr>
<td>10</td>
<td>58</td>
<td>M</td>
</tr>
<tr>
<td>Mean±SD</td>
<td>57±10</td>
<td>70% M</td>
</tr>
</tbody>
</table>

CCC, curvilinear continuous capsulorhexis; FLACS, femtosecond laser-assisted capsulotomy; SLC, selective laser capsulotomy.

### Table 2  Capsulotomy extensions, perimeters and loads for each pairwise study

<table>
<thead>
<tr>
<th>Study 1: SLC versus CCC</th>
<th>Study 2: CCC versus FLACS</th>
<th>Study 3: FLACS versus SLC</th>
</tr>
</thead>
<tbody>
<tr>
<td>Pair number</td>
<td>Extension (mm)</td>
<td>Perimeter (mm)</td>
</tr>
<tr>
<td>1</td>
<td>7±1</td>
<td>29±2</td>
</tr>
<tr>
<td>2</td>
<td>5±1</td>
<td>25±2</td>
</tr>
<tr>
<td>3</td>
<td>24±2</td>
<td>53±22</td>
</tr>
<tr>
<td>4</td>
<td>23±2</td>
<td>46±24</td>
</tr>
</tbody>
</table>

CCC, curvilinear continuous capsulorhexis; FLACS, femtosecond laser-assisted capsulotomy; SLC, selective laser capsulotomy.
statistically significantly higher for both parameters of perimeter stretch and load (p ≤ 0.01) (Figure 1).

In Study group 2 where CCC was compared with FLACS for perimeter stretch 70% of the 10 pairs demonstrate that the CCCs exceeded that of the FLACS with pairs 11, 12 and 13 being the exceptions (Figure 2A). For the load, all 10 pairs for CCC exceeded those of the FLACS. The Wilcoxon Signed Ranks Test statistical analysis resulted in an inconclusive statistical significance (p value = 0.2) when CCC was tested for superiority over FLACS with respect to perimeter extension. However, analysis of the load dataset demonstrated statistically significant superiority with the CCC requiring more load compared with FLACS to create an anterior tear, p value < 0.01 (Figure 2B).

In Study group 3 where FLACS was compared with SLC, all 10 pairs demonstrated statistically significant superiority for SLC for both increased perimeter stretch and load to tear (p ≤ 0.01) (Figure 3).

In summary, the pairwise studies demonstrated a statistically significant increase in perimeter stretch and load to tear for SLC over both CCC and FLACS. Furthermore, CCC required statistically significant more load than FLACS to tear.

Scanning electron microscopy
Images of SEM are shown in Figure 4. SLC (Figure 4A) revealed folding of the anterior capsule edge anteriorly. This appears to be from shrinkage of the anterior capsule and from local heat generation from laser energy absorption of the trypan blue stained capsule.

SEM of CCC (Figure 4B) showed a clean-cut edge revealing the fibrillar collagen structure of the capsule. The femtosecond laser capsulotomy SEM showed a irregular edge with micro-undulations resulting from repeated laser pulses with some scattered pulses as evidenced by small holes adjacent to the capsular edge (Figure 4C).

DISCUSSION
Curvilinear capsulorhexis introduced in 1990 by Gimbel and Nuehann was a revolutionary development in cataract surgery and led to considerable improvement in safety and progress in small incision phacoemulsification along with improved refractive predictability with improved lens stability. However, anterior capsule tear-outs can occur during performance of the procedure (primary anterior capsule tear) from a variety of reasons including a shallow chamber, the early learning curve of a cataract surgeon or in complex situations like an intumescent cataract. This can lead to posterior extension and involvement of the posterior segment with vitreous prolapse. Anterior radial tears can also occur during the process of cataract surgery (secondary anterior capsule tear), from for instance prolapse of a large hard nucleus during hydrodissection, instrument contact and even lens haptic contact and in turn also lead to posterior extension. The rate of anterior capsule tear has been reported to be between 0.79% and 5.5%,8 9 13 14 Posterior extension from an anterior capsule tear has been reported to be between 24%8 and 49%.9 A strong relatively tear resistant capsulotomy is therefore highly desirable.

The ideal capsulotomy in addition to being relatively strong, should be reproducible in size and also circular and central in order to adequately overlap the optic edge of the intraocular lens implant avoiding anterior optic capture with possible anterior displacement and tilt which could lead to refractive error. This has been the thrust of development of femtosecond lasers,
radio frequency capsulotomies and SLC using a non-pulsatile dye absorbing laser. Circularity and centration of the femtosecond laser has been well established and there have been reports of equivalent strength to CCC by comparing the breaking force on human anterior capsule remnants removed at surgery. Previous studies on porcine eyes have shown greater strength of FLACS over CCC while another showed FLACS was weaker than CCC. The rough capsular edge and discontinuity with tags of some FLACS capsulotomies has been attributed to potential weakness and predisposition to anterior tears. Several measures to improve capsule edge smoothness have been investigated including increasing vertical spot separation. Additionally capsule openings created with higher energy levels were in porcine eyes found to be weaker than those created with intermediate to lower energy levels. Furthermore the study by Williams et al demonstrated also in porcine eyes that larger capsules were significantly stronger, consistent with a human eye study.

This study compared three methods of anterior capsulotomy, conventional CCC, femtosecond laser capsulotomy and a new development SLC using a non-pulsatile dye absorbing laser (CAPSULaser, Excel-Lens, Los Gatos, California, USA). Unlike previous studies which used porcine eyes, this study used human cadaveric eyes which were compared pairwise and equally matched for age and cadaver time between each study group. The study also involved eliminating a pair of eyes where circularity and dimension criteria were not met as both these have been shown to influence extension and resistance to tear. The study revealed consistency of values between study groups for each type of capsulotomy (table) and this was achieved by reducing influencing variables and strictly controlling the use of eyes based on meeting dimension and circularity criteria. SLC was statistically more resistant to tear with a ratio of 146% compared with CCC. A result which was unexpected considering previous laboratory reports was the finding that CCC was statistically significantly more resistant to tear than FLACS where the ratio of FLACS to CCC was 78%. This latter finding differs from the study by Thompson et al, which showed equivalence between CCC and FLACS, however the authors did not control for variables of circularity and dimensions with considerable variation in loads from eye to eye using the same capsulotomy method. Their study also had smaller numbers of eyes (six in number) in the group comparing CCC to femtosecond laser capsulotomy which may have been a factor in not achieving statistical significance. Previous reports showing greater strength in FLACS were conducted in porcine eye studies which are known to have different anatomical thickness and elastic properties from human eyes accounting for the different findings in this human eye study.

In this study trypan blue stain was used in the SLC sub-group where a dye was required for uptake of energy from the non-pulsatile laser. There has been controversy over the impact of trypan blue stain on capsule fragility with Dick et al demonstrating increased fragility and Jaber et al later demonstrating no influence. If trypan blue does indeed increase fragility, the capsule edge of the SLC group was still statistically significant more resistant to tear than both CCC and FLACS.

SEM of the CCC and FLACS was similar to that previously reported with a very smooth anterior capsular edge with no irregularity on CCC, whereas FLACS showed considerable irregularity of the anterior capsular edge with valleys and grooves with micro- undulations and a postage stamp like appearance reflective of the repetitive pulsatile laser disruption.

Figure 3 Pairwise comparison of FLACS (diagonal patterned bars) with respect to SLC (grey bars) for (A) perimeters stretch and (B) load.

Figure 4 Scanning electron microscopy of selective laser capsulotomy (4A), continuous curvilinear capsulorhexis (4B) and femtosecond laser-assisted capsulotomy (4C).
of the capsule edge. SLC SEM on the other hand, demonstrated anterior folding of anterior capsule resulting in the capsule edge at the convexity of the fold (figure 4A). Additionally there was contracture of collagen resulting from laser energy absorption and localised thermal change. The finding of a statistically significant increased resistance to tear of SLC compared with both CCC and FLACS can be accounted by several factors including: (1) doubling of the capsular edge thickness; (2) the rolled-over folded edge which itself increases mechanical strength; (3) a smooth edge devoid of irregularity and defect, unlike FLACS in particular which had a series of micro-undulations (postage stamp appearance) and adjacent aberrant holes from pulses associated with multiple passes; both of which could promote fracture reducing resistance to tear; and (4) thermal change of the anterior capsule changing regular ordered Collagen Type IV to amorphous collagen with increased elasticity. The latter permits increased distension at a lower load before reaching the point where the capsule stretches almost maximally and in a non-linear manner ultimately leading to a tear.

Hycrodissection during phacoemulsification can lead to prolapse of a lens nucleus during surgery potentially leading to an anterior capsule tear.8 9 The distension of the capsulotomy perimeter for SLC in this study was found to average 28 mm compared with 24 mm for CCC and 23 mm for FLACS. For a 5.0 mm capsulotomy SLC would stretch 78%–85% to 8.9–9.25 mm and for both CCC and FLACS would stretch 46%–59% to 7.3–7.95 mm (table 3). This would theoretically result in less chance of an anterior capsule tear during lens prolapse and supported by the pairwise analysis of perimeters resulting in SLC being superior to, and more resistant to anterior tears than, CCC and FLACS (p<0.01).

In conclusion, this pairwise study of human cadaver eyes controlled for deviations in circularity and dimensional size, comparing three methods of capsulotomy demonstrated SLC was significantly stronger than curvilinear capsulorrhexis which in turn was significantly stronger than femtosecond laser capsulorrhexis. SEM findings demonstrated structural changes supporting these findings. Clinically SLC may provide increased surgical safety by reducing the chances of anterior capsular tears during cataract surgery.

### Contributors
SD wrote the manuscript. DHM was involved in study design, data and statistic analysis. S-PC and S-ET performed the experiments. RP made written contributions to portions of the manuscript.

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### Competing interests
SD is a consultant and medical monitor to Bausch and Lomb manufacturer of the Victus laser used in this study. He is also an equity shareholder in Excel-Lens, manufacturer of the CAPSULaser device studied. RP and DHM are equity shareholders in Excel-Lens, manufacturer of the CAPSULaser device studied. Dr S-PC is a speaker for Bausch and Lomb.