Performing Reliable Lens Capsulotomy in the Presence of Corneal Edema With a Femtosecond Laser

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Endothelial dysfunction caused by conditions such as Fuchs’ endothelial corneal dystrophy (FECD) and bullous keratopathy result in visual impairment due to corneal edema and scarring.1–3 When corneal opacification occurs in addition to corneal edema, optimized visualization of the lens capsule by capsular staining or retroillumination may assist continuous curvilinear capsulorhexis (CCC),5,6 but an endothelial transplant is usually indicated at the same sitting. Surgical management of corneal opacification is usually indicated at the same sitting. Surgical management of cataract disease is achieved by selective tissue transplantation by Descemet stripping automated (DSEA) or Descemet membrane endothelial keratoplasty (DMEK).7,8 These interventions are usually undertaken following or combined with removal of cataract.7,9

Cataract surgery may contribute to corneal decompensation because of endothelial cell damage from phacoemulsification energy, however corneal edema may already be apparent. Central corneal thickness increases from healthy individuals (558 μm) incrementally from 586 μm in grade 1 FECD to 648 μm in grade 6 disease.10 However, the precise thickness that predicts the likelihood of decompensation during phacoemulsification in FECD is ill defined. Relatively thin corneas (such as those with grade 1 disease < 600 μm) may be relatively less edematous (thickened), despite frank evidence of decompensation and yet a greater central corneal thickness (CCT) may be seen in an optically clear cornea (e.g., greater than 640 μm following pre-existing high CCT).11

Femtosecond laser-assisted cataract surgery (FLACS) offers a potentially more consistent way of undertaking capsulotomy by creating a semiautomated anterior lens capsulotomy.12–15 The effects of a thickened yet transparent cornea on successful FLACS is unknown. There is potential to use this technology to reduce disruption of endothelial cells by femto-fragmentation. This may have a specific role in cases where the endothelium is at high risk such as FECD.16,17 Despite these advantages, femtosecond laser firing patterns are vulnerable to enhance light scatter at deeper dissections, even in optically clear corneas. Increased variability is seen, for example, in LASIK flap creation in the peripheral (thicker) compared with central (thinner) cornea.18 This could potentially make femtosecond tissue cutting more challenging in the presence of corneal edema (e.g., penetrating keratoplasty).19,20 As FLACS relies on an optically clear interface, the potential advantage of laser
capsulotomy and fragmentation could hypothetically be negated by the inability of the laser to fire successfully through a thickened cornea.

FLACS capsulotomy may induce capsular tags, leading to more serious complications such as anterior capsular tears.\(^1\)\(^{-}\)\(^{23}\) The strength of the capsulotomy in resisting rupture during manipulation of lens material is critical to success.\(^24\)\(^{-}\)\(^{29}\) It has recently been shown that increasing the laser energy pulse may adversely affect the strength of the lens capsulotomy.\(^{23}\) This effect was noticeable with a high-energy FLACS (‘microjoule energy’) platform. We have previously shown however that in a low-energy high-frequency system (operating in the nanojoule range) that a safe, smooth capsulotomy could be achieved.\(^{30}\) The utility of such a system in the presence of corneal edema is currently unknown, but microjoule energy platforms may penetrate edematous corneas more effectively. High-energy systems have been shown to induce increased apoptosis of the lens epithelium during femtosecond capsulotomy, however, suggesting an advantage for manual capsulotomy including minimal wound healing reaction and lens capsule cell death.\(^{31\)\(^{-}33}\) By contrast, suction trephination during penetrating keratoplasty demonstrates less apoptosis than femtosecond laser.”\(^{34}\) The potential for safely increasing energy in nanojoule steps to overcome corneal edema has yet to be resolved.

The aim of this study was to determine the efficacy of the low-energy Ziemer LDV Z8 liquid interface femtosecond laser platform during capsulotomy in the context of corneal edema and at different energy settings to compensate for thickened corneas.

### METHODS

#### Porcine Capsules

Ex vivo cadaveric porcine eyes were sourced from a local abattoir, and capsulotomies performed at less than 6 hours or greater than 24 hours post enucleation and evaluated with the Ziemer LDV Z8 femtosecond laser (software version ×5054; Ziemer, Port, Switzerland) for different laser energies (5-mm diameter, 0.8-mm height, cut speed 50 mm/s, energy 90%–150%), as previously described.\(^{30\)\(^{-}35}\) Briefly, due to a tendency to cadaveric porcine corneal epithelium to slough, for consistency the corneal epithelium was debrided and the globe mounted in a suction stand prior to docking with a liquid patient interface followed by suction, liquid immersion, and attachment of the laser head. Corneas with impairment of subjective clarity (i.e., obvious scarring) opacities were excluded. Forty-four globes were used with n = 6 in each group for further experimentation (Table)

Lens capsulotomy strength was determined by the single column universal testing system (3343; Instron Corp., Canton, MA, USA) after removal of the lens en bloc with nucleus intact to follow expression with an ophthalmic viscoelastic device (Viscoat, Alcon, Fort Worth, TX, USA). Two mushroom-shaped pins were placed posterior to the capsulotomy edge and the rate of pin displacement was set at 6 mm/min. Resistance of the capsulotomy to rupture was measured in meganewtons and the stretching ratio by (capsulotomy size mm + displacement mm)/capsulotomy size mm, as previously described.

#### Human Capsules

Increased cell death at the capsule edge may potentially correlate with a less smooth capsule edge morphology.\(^{31}\) In order to determine cell death at increasing energy during FLACS capsulotomy, we evaluated lens capsules ex vivo following routine cataract surgery. Human capsules were collected during routine phacoemulsification cataract surgery (MA, JSM) following informed consent and institutional review board approval in keeping with the tenets of the Declaration of Helsinki (CIRB Ref 2015/2565, Singapore). All patients’ corneal thickness were normal, in the range of 520 to 550 μm. Manual CCC was achieved with a 27G needle with Viscoat ophthalmic viscoelastic device (Alcon) or capsulotomy with the Ziemer LDV Z8 (Ziemer) femtosecond laser (software version ×5054) for different laser energies (5-mm diameter, 0.8-mm height, cut speed 50 mm/s, energy 90%–150%), as previously described.\(^{30\)\(^{-}35}\) Mean patient ages (n = 6 for each group) were: manual 69 years (range, 49–75), 90% energy 74 years (56–85), 130% energy 71 years (56–76), and 150% 68 years (57–72) (P = 0.62). All cataracts were senile in origin and with no known trauma or coexisting disease.

After anterior capsule extraction, all specimens were immediately fixed in freshly prepared neutral buffered 4% paraformaldehyde in PBS (0.01M; First Base, Singapore) for 1 hour at 4°C. During fixation, the samples were carefully placed with epithelial side facing upward to prevent any mechanical damage to cells that might induce unnecessary apoptosis. After...
femtosecond laser at low energy. 15,35,37 We have previously evaluated the role of the Ziemer LDV Z8 nanojoule energy platform on lens capsulotomies, which had shown consistency and resistance to rupture with this system. 36 Increasing energy in the microjoule range may adversely affect capsulotomy strength and increase cell death in high-energy platforms (5–15 μJ). 25,31 But smaller increase in energy as a means of safely circumventing the effects of corneal edema with a low-energy laser (<100 nJ or 0.1 μJ) 15 has not been previously determined. Therefore, in this study we found that the consistency of circularity and lens elasticity was seen both in clear corneas and at increased laser energy—from a conventional recommended setting of 90% to 150%. These observations were similar to our previous experimentation with porcine and human lens capsulotomies.
at 5-mm diameter at 90% energy. However, in the presence of corneal edema, a successful lens capsulotomy was only achieved at increased energy settings (150%). The lower energy setting resulted in 50% failure in edematous corneas including irregular or incomplete cuts, which could lead to anterior capsular tears. This represents a potential challenge for patients who may have corneal edema associated with FECD undergoing FLACS.

The laser focus is anatomically more posterior to facilitate capsulotomy and successful cuts demonstrated that the edges of the capsulotomy appeared smooth and consistent across energy settings, despite edema. We have previously shown similar smooth capsulotomy edge morphology with the LDV Z8 using nanojoule energy, however, there has been shown to be significant variation in the appearance among microjoule energy laser platforms, especially when increasing the energy settings from 5 to 15 μJ. These changes include serrations and morphologic changes including coagulation of collagen fibers. Although increasing energy within one of the microjoule energy laser platforms (e.g., LenSx) showed increased disruption to capsule edge morphology, considerable variation exists in other platforms (e.g., Catalys, Victus)
Figure 2. The effects of energy alterations on capsulotomy circularity, edge, and strength in a nonedematous cornea. Representative images at increasing energy at less than 6 hours post enucleation with circularity (A). Predicted and actual capsule sizes are shown in (B). Scanning electron microscopy images of capsule edge taken at increasing energy are shown in (C). Lenses were removed with nucleus intact and resistance of the capsulotomy to rupture was measured in millinewtons and the stretching ratio by (size mm + displacement mm)/size. Two mushroom-shaped pins were placed posterior to the capsulotomy edge and the rate of pin displacement was set at 6 mm/min. Comparison between strength and stretch ratio at different energies are shown in (D). *P < 0.05. **P < 0.01.
Figure 3. The effects of energy alterations on capsulotomy circularity, edge, and strength in a edematous cornea. Representative images at increasing energy at greater than 24 hours post enucleation with circularity (A). Predicted and actual capsule sizes are shown in (B). Scanning electron microscopy images of capsule edge taken at increasing energy are shown in (C). Lenses were removed with nucleus intact and resistance of the capsulotomy to rupture was measured in millinewtons and the stretching ratio by (size mm + displacement mm)/size. Two mushroom-shaped pins were placed posterior to the capsulotomy edge and the rate of pin displacement was set at 6 mm/min. Comparison between strength and stretch ratio at different energies are shown in (D). NS, not significant. *P < 0.05. **P < 0.01.
Figure 4. Human lens capsule TUNEL staining following femtosecond capsulotomy. Positive controls pretreated with DNase and TdT enzyme and negative controls without TdT enzyme are shown in (A). Representative fluorescence microscopy images (×10) from manual cut and LDV Z8 femtosecond cut capsulotomies at 90%, 130%, and 150% are shown in (B). Percentage TUNEL-positive cells (taken from 6 randomized regions along the cutting edge of each sample in a rectangular 100-μm field) are shown in (C). *P < 0.05.
reflecting incrementally larger differences in energy delivery.21,23,31,38,39 We did not see this with the nanojoule energy laser used in clear, nonthickened, or edematous corneas in this study.

It is likely that the magnitude of the energy increase with a nanojoule energy platform (90% vs. 130% vs. 150%) does not induce collagen melting and denaturation seen at higher energy settings with a microjoule energy platform.25 Unlike previous studies evaluating increasing microjoule energy,25 we saw no significant difference in the strength of the capsulotomy at increasing energy in the nanojoule range (90% vs. 150% vs. 150%). This was unaffected by the presence or absence of corneal edema and consistent with the edge morphology demonstrated. This suggests that the 150% setting has incrementally lower energy than for platforms operating in the microjoule range.15 We cannot explain why the stretch ratio increased at 150% energy as intuitively as we might have expected this to reduce. The elastic recoil seen in the capsulotomy was consistent with our previous data and the behavior of porcine lenses.40 It would suggest that this was unaffected with the LDV Z8 laser and despite the personal observation of a temporary whitening of the capsulotomy edge at higher (150%) energy setting in patients (MA) compared with 90% energy.

TUNEL staining demonstrated low apoptosis across energy settings. DNase-treated positive controls showed 100% apoptosis; while negative control without TdT enzyme had 0%. This validated the assay and is important when considering energy disruption to the capsule itself, which was low, albeit at a statistically higher level in the 150% setting. It is unclear why this peaked at this energy level but it is possible that removal of the lens capsules may have been more traumatic in these capsules. At less than 2% TUNEL-positive cells however we postulate that this appeared to be clinically insignificant. Mayer et al.31 previously showed that by contrast there were significantly higher levels of cell death in the lens capsule at increasing energy, from 30 cells/mm² at 5 μJ to greater than 80 cells/mm² at 15 μJ laser pulse energy. Due to the possibility of underrepresenting TUNEL-positive cells by counting three randomized regions of the cutting edge,31 we quantified percentage of cell death along the whole edge and suggest that it is more representative than cells per millimeter squared. Nonetheless, counts were 7.5 cells/mm² for manual cut versus 13 cells/mm² at 90% energy versus 68 cells/mm² at 130% energy versus 42 cells/mm² at 150% with the nanojoule laser. Even at 150% the nanojoule energy laser was focused with minimal collateral damage, showing the safety of increasing the energy setting in the context of corneal edema.

Limitations in this study include the absence of data on corneal guttae and/or scarring.31,32 The results are therefore more relevant for patients who have FEDC with the presence of corneal thickening. In conditions such as FEDC or viral endothelitis where guttatae and/or scarring may coexist the influence of these parameters on laser scatter are unknown. Correlation between and porcine and human corneal edema is expected this to reduce. The elastic recoil seen in the capsulotomy was consistent with our previous data and the behavior of porcine lenses.40 It would suggest that this was unaffected with the LDV Z8 laser and despite the personal observation of a temporary whitening of the capsulotomy edge at higher (150%) energy setting in patients (MA) compared with 90% energy.

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We have demonstrated that the LDV Z8 femtosecond laser was able to create a consistent, circular, and smooth capsulotomy that was capable of resisting stress at increased energy. Because of the low energy delivered by the Ziemer Z8 platform, even the incremental increases in energy appeared to have minimal effect on lens capsule morphology and strength and negligible influence on cell death. In eyes with FECD and early decompensation, there are potential benefits of undertaking FLACS, and thereby reducing endothelial damage from phacoemulsification energy.41,42 Increasing energy in the nanojoule range appears to enhance consistency in the ability to complete a capsulotomy in an edematous cornea, which may be useful when performing FLACS in eyes with concurrent corneal pathology such as FEDC. We therefore advocate that greater energy should be used in an edematous cornea for FLACS-assisted capsulotomy.

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References

Femtosecond Laser to Overcome Corneal Edema


