Biocompatibility of intraocular lens power adjustment using a femtosecond laser in a rabbit model

Liliana Werner, MD, PhD, Jason Ludlow, MD, Jason Nguyen, MD, Joah Aliancy, MD, Larry Ha, BS, Bryan Masino, BS, Sean Enright, BS, Ray K. Alley, BS, Ruth Sahler, MSc, Nick Mamalis, MD

Purpose: To evaluate the biocompatibility (uveal and capsular) of intraocular lens (IOL) power adjustment by a femtosecond laser obtained through increased hydrophilicity of targeted areas within the optic, creating the ability to build a refractive-index shaping lens within an existing IOL.

Setting: John A. Moran Eye Center, University of Utah, Salt Lake City, Utah, USA.

Design: Experimental study.

Methods: Six rabbits had phacoemulsification with bilateral implantation of a commercially available hydrophobic acrylic IOL. The postoperative power adjustment was performed 2 weeks after implantation in 1 eye of each rabbit. The animals were followed clinically for an additional 2 weeks and then killed humanely. Their globes were enucleated and bisected coronally just anterior to the equator for gross examination from the Miyake-Apple view to assess capsular bag opacification. After IOL explantation for power measurements, the globes were sectioned and processed for standard histopathology.

Results: Slitlamp examinations performed after the laser treatments showed the formation of small gas bubbles behind the lenses that disappeared within a few hours. No postoperative inflammation or toxicity was observed in the treated eyes, and postoperative outcomes and histopathological examination results were similar to those in untreated eyes. The power measurements showed that the change in power obtained was consistent and within ±0.1 diopter of the target.

Conclusions: Consistent and precise power changes can be induced in the optic of commercially available IOLs in vivo by using a femtosecond laser to create a refractive-index shaping lens. The laser treatment of the IOLs was biocompatible.


Studies have shown that despite the many advances in cataract surgery, incorrect intraocular lens (IOL) power remains 1 of the most frequent causes of IOL exchange.1–3 A We recently published an overview of the adjustable IOL technologies that are available or under development, which could be used to mitigate the problem of incorrect IOL power.4 These include IOL technologies that can be adjusted using secondary surgical procedures and IOLs that can be adjusted noninvasively in the postoperative setting. Calhoun Vision’s light-adjustable IOL is the noninvasive adjustable IOL closest to commercial availability in the United States because it is undergoing the third and final phase of U.S. Food and Drug Administration clinical trials.5,6 Among other noninvasive adjustable IOL technologies under development discussed in our review paper is refractive-index shaping using the femtosecond laser.4

Perfect Lens LLC has developed a femtosecond laser system for IOL power adjustment based on the concept of refractive-index shaping. The system uses green light (520 nm) and operates with energy levels that are below the threshold for ablation or cuts. Intraocular lens power changes are obtained through a laser-induced chemical reaction in a targeted area of the IOL optic substance, with a
localized increase in hydrophilicity and decrease in the refractive index. Simultaneous with these changes, the laser builds a refractive-index shaping lens within the targeted area. Studies using hydrophobic and hydrophilic acrylic IOLs have shown the consistency and precision of the power changes that can be induced in the optic of commercially available IOLs in vitro. To our knowledge, the current study is the first to evaluate the biocompatibility and efficacy of this technology in vivo, using the rabbit model.

MATERIALS AND METHODS

Six New Zealand white female rabbits weighing 2.8 to 3.2 kg were acquired from approved vendors in accordance with the requirements of the Animal Welfare Act for use in this study. All rabbits were treated in accordance with guidelines set forth by the Association for Research in Vision and Ophthalmology and the Animal Welfare Act regulations as well as the Guide for the Care and Use of Laboratory Animals.

Each animal was prepared for surgery by pupil dilation with cyclopentolate hydrochloride 1.0% and phenylephrine 2.5% drops, as described in previous studies. Anesthesia was obtained with an intramuscular injection of ketamine hydrochloride (50 mg/kg) and xylazine (7 mg/Kg) in a mixture of 7:1, respectively. All surgeries were performed by the same surgeon (N.M.). Using aseptic technique and a surgical microscope, a fornix-based conjunctival flap was fashioned. A 3.0 mm limbal incision was made using a 3.0 mm keratome, and sodium hyaluronate 1.6% (Amvisc Plus) was injected intracameraly. A capsulorhexis forceps was used to create a well-centered continuous curvilinear capsulotomy with a diameter aimed at 5.5 mm. After hydrodissection, the phaco handpiece (Infiniti System, Alcon Laboratories, Inc.) was inserted into the posterior chamber for removal of the lens nucleus and cortical material. One milliliter of epinephrine 1:1000 and 0.5 mL of heparin (10 000 USP units/mL) were added to each 500 mL of irrigation solution to facilitate pupil dilation and control inflammation. The endocapsular technique was used with the phacoemulsification to take place entirely within the capsular bag. The residual cortex was then removed by irrigation/aspiration. The same ophthalmic viscosurgical device was used to inflate the capsular bag, and a single-piece hydrophobic acrylic preloaded yellow IOL (CT Lucia 601PY, Carl Zeiss Meditec AG) was then injected in the capsular bag. All IOLs used in the study had the same labeled power. Wound closure was achieved with a 10-0 monofilament nylon suture after removal of OVD.

An ointment combination (neomycin/polymyxin-B sulfates and dexamethasone) was applied to the eyes after the surgery was performed, and the ointment was used 4 times a day for the first postoperative week. In the second postoperative week, each animal received topical prednisolone acetate drops 4 times per day.

The eyes of the rabbits were evaluated by slitlamp examination and scored for ocular inflammatory response weekly after pupil dilation. A standard scoring method in 11 categories was used at each examination, including assessment of corneal edema and the presence of cell and flare within the anterior chamber. Retroillumination images with the dilated pupil were obtained for photographic documentation regarding inflammatory reactions, anterior capsule opacification (ACO), posterior capsule opacification (PCO), and observed capsule fibrosis. The ACO at the area of the anterior capsule contacting the anterior optic surface was scored from 0 to 4. The PCO behind the IOL optic was scored from 0 to 4.

Intraocular Lens Power Adjustment by Laser

Postoperative IOL power adjustment was performed 2 weeks after IOL implantation in only 1 of the eyes, and the rabbits were followed clinically for an additional 2 weeks. The eye to receive the power adjustment was selected as a function of the clarity of the capsular bag in front of the lens (no or minimum proliferation or pearl formation in front of the lens, no or minimum ACO or capsulorhexis phimosis). For the laser adjustment, each animal was prepared by pupil dilation and anesthesia as done for the surgical implantation procedure. The rabbit was placed horizontally on a support/bed constructed with a 3-dimension printer (allowing rotation/tilt of the animal in different directions), with the designated eye facing up to allow the connection to the patient interface (Figure 1, A). The interface was purpose-designed for the rabbit eye based on measurements described in a previous study. Alignment of the rabbit eye and docking to the laser system through the interface were then performed under the control of the video and optical coherence tomography (OCT) systems of the laser device, with subsequent laser treatment targeted at a +3.6 diopter (D) power change. A forceps was used to displace the nictitating membrane of the rabbit eye immediately before docking. Slitlamp examination of the eyes was performed immediately after laser treatment and at different timepoints after treatment.

Pathology

After the final clinical examination 4 weeks postoperatively, the animals were anesthetized and humanely killed with a 1 mL...
intravenous injection of pentobarbital sodium/phenytoin sodium. The globes were enucleated and placed in 10% neutral buffered formalin. They were then bisected coronally just anterior to the equator. Gross examination from the posterior aspect (Miyake-Apple view) was performed to assess ACO and PCO development. The ACO at the area of anterior capsule contacting the anterior optic surface was scored from 0 to 4. Central PCO related to the central 3.0 mm behind the optic was scored from 0 to 4. Peripheral PCO related to the peripheral area behind the optic was scored from 0 to 4. Soemmerring ring formation related to proliferative material within the equatorial region of the capsular bag, outside of the optic, had a score of intensity from 0 to 4 and a score of area related to the number of quadrants involving the highest intensity.

The IOLs were then carefully removed from the capsular bag of each eye (treated and untreated IOLs). Proliferative material attached to the IOLs was removed using surgical sponges, and the IOLs were immersed in vials containing distilled water. Light microscopy was then performed at room temperature to evaluate the explanted IOLs, and photomicrographs were taken with a camera coupled to the light microscope. The IOLs were replaced in the vials containing distilled water and were forwarded to Perfect Lens LLC for power measurements. All measurements were taken using the PMTF device (Lambda-X S.A.), a power and modulation transfer function (MTF) measurement device designed for refractive and diffractive IOLs, which is ISO 11979-2 compliant; it has an ISO 11979-2 model eye and uses a measurement wavelength of 546 nm. The globes were sectioned and the anterior segments, including any remaining capsular bags, were processed for standard light microscopy and stained with hematoxylin–eosin. The histopathological analyses focused on signs of inflammatory reaction or toxicity in the different structures of the anterior segment of the eyes.

Opacification data were analyzed using Excel software (Microsoft Corp.).

RESULTS
Overall, all implantation procedures were uneventful and the IOLs could be fully injected within the capsular bag. At the 1-week examination, nearly all operated eyes had a mild inflammatory reaction with fibrin in front of the lens or at the level of the capsulorhexis edge. Fibrin formation had completely resolved by the 2-week examination, when a mild amount of PCO started to be observed in nearly all eyes. Most eyes at this timepoint also had proliferative lens cortical material or pearl formation in front of the IOL.

All laser power adjustment procedures were also uneventful, and the duration of the laser treatment per se was fast (23 seconds). Under slitlamp examination, the phase-wrapped structure created by the laser could be observed within the optic substance of all treated IOLs. The examination also showed the formation of gas bubbles.
between the posterior surface of the IOL and the posterior capsule, which disappeared within 5 hours (Figure 2). Other observations included mild corneal edema and conjunctival injection, which could be related to the eye remaining open during the alignment step of the procedure. No aqueous flare, cells, iris hyperemia, or fibrin formations were observed at any of the post-laser slitlamp examinations, and the process did not create glistenings in the IOLs.

By the 3-week examination, most eyes with pearl formation in front of the lens had developed posterior synechia formation in 1 quadrant. The PCO formation progressively increased in intensity throughout the clinical follow-up (Figure 3). At the 4-week examination, the mean PCO score was 2.25 ± 0.68 (SD) in the laser-treated eyes and 2.91 ± 0.66 in the untreated eyes (2-tailed \( P = .06; t \) test paired 2 sample for means). The ACO was observed in all eyes (usually as a fibrotic rim at the level of the capsulorhexis edge), and 2 untreated eyes developed capsulorhexis phimosis.

Under gross examination from the Miyake-Apple view of the anterior segments of the enucleated eyes, all the IOLs were found to be symmetrically fixated within the capsular bag and overall centered in relation to the ciliary processes (Figure 4). Capsular bag opacification was scored as follows: The mean central PCO score was 1.5 ± 1.0 in the treated eyes and 2.0 ± 0.63 in the untreated eyes (2-tailed \( P = .27; t \) test paired 2 sample for means). The mean peripheral PCO score was 2.33 ± 0.81 and 2.5 ± 0.54, respectively (2-tailed \( P = .61; t \) test paired 2 sample for means). The mean Soemmerring ring formation (intensity X area) score was 8.33 ± 0.51 in the treated eyes and 8.0 ± 0 in the untreated eyes (2-tailed \( P = .17; t \) test paired 2 sample for means). Clinical and gross postmortem evaluation showed no significant differences in the parameters assessed between the study eyes and the control eyes.

Light microscopy of the explanted IOLs showed the phase-wrapped structure created by the laser within the optic substance in all treated IOLs. The phase-wrapped structure was mildly decentered in some of the IOLs. Small amounts of proliferative material were also found on the surface of most of the explants. No IOLs showed the presence of damage, deformation, pitting, or marks (Figure 5). The mean refractive-index shaping lens dioptr change was measured after full hydration of the explanted IOLs. The mean dioptr difference between the refractive-index shaping dioptr and the control lens dioptr was +3.58 ± 0.26 D (Table 1). The change in power obtained was consistent, and the mean was within ±0.1 D of the target.

Examination of multiple histopathological sections cut from each eye under the light microscope showed that there was no untoward toxicity or inflammation in the eyes that had laser treatment of the IOLs or in the control eyes (Figure 6).

**DISCUSSION**

The principles of the technology allowing creation of a refractive lens within an existing IOL using a femtosecond laser have been recently described. A significant negative refractive-index change in a hydrophobic acrylic material is created by exposing such material to water and light of a particular wavelength and pulse energy of a femtosecond laser. The laser creates an increase in hydrophilicity in targeted areas within the hydrophobic material. The treated area of the material then absorbs water and thus reduces its refractive index. In a hydrophilic acrylic material, the targeted area is rendered more hydrophilic. After laser treatment, the surface of the IOL and its untreated substance remain unchanged. The laser then builds a refractive-index shaping lens within the targeted area of the lens optic substance, which is rendered more hydrophilic. To create the refractive-index shaping lens within the IOL, one cannot simply create a traditional convex or concave lens within the lens optic. A significant refractive change in such a small area can only be obtained with a phase-wrapped structure that contains the entire curvature of a traditional convex or concave lens collapsed into 1 layer. The phase-wrapped lens is a theoretically perfect Fresnel lens. A refractive-index change (\( \Delta n \)) of 0.01 in a 6.0 mm conventional lens with a height of 200 \( \mu \)m will
produce a diopter change of 0.4 D, whereas a 0.01 Δn in a phase-wrapped lens with the same size lens creates a diopter change of 3.3 D.7

A recent study8 assessed the chemical basis for the alteration of the refractive properties of an acrylic IOL with a femtosecond laser. In that study, the acrylic material was tested by several microscopic methodologies, including laser-induced fluorescence microscopy, Raman microscopy, and coherent antistokes Raman scattering microscopy, to determine the nature of the changes created in the material by the exposure to the femtosecond laser. The authors found photo-induced hydrolysis of the polymeric material in aqueous media, which produced 2 hydrophilic functional groups: acid group and alcohol group. After the exposure of the polymeric material, water slowly diffused to the sites with increased hydrophilicity, forming hydrogen bonds, typically over a 24- to 72-hour period, to create a negative refractive-index change in the polymeric material. Based on the same microscopic methodologies used, no leachables were generated in the process. Also, standard leachable tests have been performed on modified IOLs and no leachables were found.8

The consistency and precision of the power changes induced by the laser have been shown in vitro. Another recent study7 found that the refractive-index change altered the dioptic power of commercially available yellow hydrophobic acrylic IOLs to within ±0.1 D of the targeted change without a significant reduction in the MTF. A more recent study performed in our laboratoryC also showed the consistency and precision of the power change by this technology in commercially available hydrophobic acrylic lenses with and without a blue-light filter, without inducing significant changes in IOL light transmission.

Our current in vivo study confirmed that postoperative outcomes in terms of uveal and capsular biocompatibility were similar between treated lenses and untreated lenses, as shown during clinical examination and by complete histopathology. The laser power adjustment procedure did not induce inflammatory reactions in the eye or damage to the IOL optic. Alignment of the rabbit eye under the laser system for the adjustment procedure was challenging because it was necessary to anesthetize the animal, which would not be the case in a clinical situation. Even though an eye interface had to be specially designed for this study, which was also the first performed in vivo, the change in power obtained was consistent in the group of treated eyes. It is noteworthy that power measurements of the IOLs were not performed before implantation in the rabbit eyes to avoid compromising the sterility of the IOLs because the main objective of the current study was to evaluate biocompatibility after laser treatment. Therefore, the method used to estimate the changes in power after laser treatment was based on measurements done with the power and MTF device after IOL explantation.

The most likely cause of postoperative refractive errors after IOL implantation is incorrect IOL calculation resulting from incorrect measurements of the eye.4 Also, current standards regarding IOL power labeling allow a tolerance of ± 0.30 D for IOLs of 0.00 D to 15.00 D or less. The tolerance increases to ± 0.40 D for IOLs with a power from greater than 15.00 D to 25.00 D or less, which means that an IOL of 22.61 D and another of 23.39 D could be labeled with a

Table 1. Power of the IOLs implanted in the rabbit eyes, measured with a PMTF device after explantation of the lenses 4 weeks postoperatively after full hydration.

<table>
<thead>
<tr>
<th>Rabbit</th>
<th>IOL Power (D)</th>
<th>Change</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>+26.5</td>
<td>+3.3</td>
</tr>
<tr>
<td>2</td>
<td>+26.9</td>
<td>+3.7</td>
</tr>
<tr>
<td>3</td>
<td>+27.0</td>
<td>+3.3</td>
</tr>
<tr>
<td>4</td>
<td>+26.7</td>
<td>+3.6</td>
</tr>
<tr>
<td>5</td>
<td>+27.0</td>
<td>+4.0</td>
</tr>
<tr>
<td>6</td>
<td>+26.8</td>
<td>+3.6</td>
</tr>
</tbody>
</table>

IOL = intraocular lens

Figure 5. Light photomicrographs of the IOLs explanted from the eyes of the same rabbit. Small amounts of proliferative material can be seen attached to the surface of the IOLs. The phase-wrapped pattern can also be seen within the substance of the treated IOL. A: Treated IOL. B: Untreated IOL.
dioptric power of 23.00 D or the IOL of 23.39 D could be labeled as both 23.0 D and 23.5 D. All these factors make postoperative IOL adjustment technologies particularly interesting.

In summary, we described what we believe to be the first in vivo study evaluating the biocompatibility of a new application of the femtosecond laser; that is, postoperative IOL power adjustment, which has potential advantages over existing IOL power adjustable technologies. It can be applied to any commercially available hydrophobic or hydrophilic acrylic IOL because the process does not depend on a special IOL material. Power adjustment is noninvasive and fast and can be performed under topical anesthesia. The dioptric power of the IOL can be increased or decreased to account for surgical errors, IOL tilt, IOL decentration, or a change in the physical characteristics of the eye. Multiple adjustments to the same IOL can be performed because each adjustment only changes a very thin layer within the IOL optic substance. Premium functions can be added to the IOL and removed later, if necessary. An added multifocal pattern can, for example, be canceled by application of a pattern with opposite characteristics. The use of special protective spectacles is not necessary after treatment, such as in Calhoun Vision’s light-adjustable IOL technology, which requires the patient to wear special spectacles until lock-in of the IOL power is complete. The laser system used for IOL power adjustment could be designed to also perform corneal and cataract surgery procedures. Further evaluation of this promising technology is warranted.

WHAT WAS KNOWN
- Refractive properties of commercially available hydrophobic or hydrophilic acrylic IOLs can be customized after implantation using a femtosecond laser through construction of a refractive-index shaping lens within the implanted IOL with micrometer precision.

WHAT THIS PAPER ADDS
- Evaluation of the refractive-index shaping lens using the femtosecond laser in vivo in the rabbit model resulted in similar uveal and capsule biocompatibility outcomes in treated eyes and untreated eyes as well as in the power change to the treated lenses with no damage to their optics.
REFERENCES


OTHER CITED MATERIAL


Disclosures: Mr. Enright, Mr. Alley, and Ms. Sahler are employees of Perfect Lens LLC. None of the other authors has a financial or proprietary interest in any material or method mentioned.

First author:
Liliana Werner, MD, PhD
Department of Ophthalmology and Visual Sciences, John A. Moran Eye Center, University of Utah, Salt Lake City, Utah, USA