The Safety, Pharmacokinetics, and Efficacy of Intraocular Celecoxib

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PURPOSE. To determine safety, pharmacokinetics, and anti-inflammatory effects of intraocular celecoxib.

METHODS. The right eye of animals was injected with 1.5, 3, or 6 mg celecoxib prepared in dimethyl sulfoxide (DMSO). Left eyes served as controls and received 0.1 mL DMSO. Electroretinograms (ERG) were obtained at baseline and at 1, 4, and 12 weeks, and eyes were enucleated afterward for histopathologic analysis. For pharmacokinetics, 3 mg celecoxib was injected, and vitreous and retina/choroid drug levels were then analyzed at specific time points. For efficacy, 1 μg lipopolysaccharide was injected to induce inflammation; the right eye was then injected with 3 mg celecoxib (six eyes) or 2 mg triamcinolone acetonide (six eyes) and the left eye with saline. Twenty-four hours later, aqueous fluid was removed, and total leukocyte concentration and prostaglandin E2 (PGE2) concentration were determined.

RESULTS. Histologic and ERG studies demonstrated no signs of retinal or optic nerve toxicity. After a single 3-mg injection, vitreous (0.06 μg/mL) and retina/choroid (132.31 μg/g) celecoxib concentrations at 8 weeks exceeded median inhibitory concentration. Treatment with celecoxib and triamcinolone significantly reduced total leukocyte count by 40% (P = 0.02) and 31% (P = 0.01), respectively. Reduction in PGE2 levels paralleled reduction in leukocyte counts (P < 0.05). There was no increase in intraocular pressure, but cataract formation was observed at higher concentrations.

CONCLUSIONS. Intraocular injection of celecoxib appeared to be nontoxic and demonstrated excellent penetration into the retina/choroid and sustained drug levels out to 8 weeks. Celecoxib demonstrated potent anti-inflammatory effects, but there was an association with cataract formation at higher doses.

Keywords: NSAIDs, prostaglandins, celecoxib, intravitreal drug delivery, inflammation

Cyclooxygenase (COX) is a critical enzyme in the inflammatory process and catalyzes the biosynthesis of proinflammatory prostaglandins (PGs) from arachidonic acid.1 Nonsteroidal anti-inflammatory drugs (NSAIDs) are potent inhibitors of COX enzymes and thereby the synthesis of all downstream PGs. Within the eye, PGs disrupt the blood–ocular barrier, increase vasodilation, and facilitate leukocyte migration. Consequently, topical formulations of NSAIDs have been shown in several well-designed clinical studies to reduce intraocular inflammation and macular edema (ME) after cataract and vitrectomtary surgery.1–3

A growing body of scientific evidence suggests that PGs may play a pathogenic role in diabetic retinopathy (DR) and age-related macular degeneration (AMD), the most frequent causes of legal blindness in working-age and elderly (>65) adults, respectively.4–6 Topical or oral application of NSAIDs, however, does not result in significant retinal drug levels.7 Intraocular administration, on the other hand, provides considerably higher retinal levels while minimizing systemic exposure.8

Celecoxib is a sulfonamide NSAID and selective COX-2 inhibitor used in the treatment of inflammatory arthritis, osteoarthritis, pain, and familial adenomatous polyposis. Celecoxib is more than 300 times more selective for COX-2 than COX-1.9 Cyclooxygenase-2 is the inducible isoform of COX and predominately involved in inflammatory responses, while COX-1 is constitutively expressed and involved in daily housekeeping functions.1 Consequently, prolonged selective inhibition of COX-2 may be preferable.

Recent evidence supports a therapeutic role of celecoxib in DR. Treatment with celecoxib in streptozotocin-induced diabetic rats reduces retinal VEGF expression and vascular leakage,10 and a prospective controlled trial demonstrated that oral celecoxib significantly reduced vascular leakage in patients with DR despite premature stoppage due to concerns regarding cardiovascular toxicity.11 Intraocular injection minimizes systemic toxicity, and because of its relative insolubility in vitreous, celecoxib possesses ideal properties for sustained intraocular drug delivery.12

Therefore, the primary intent of this study was to (1) assess the histopathologic and electroretinographic safety of escalating doses of intraocular celecoxib in rabbit eyes, (2) determine vitreous and retina/choroid celecoxib concentration over time after a single intraocular injection, and (3) investigate the anti-inflammatory effects of intraocular celecoxib in an animal model of uveitis.
Intraocular Celecoxib

METHODS

Animals

The Vanderbilt Animal Care and Use Committee, Nashville, Tennessee, approved all aspects of this investigation, and the study adhered to the ARVO Statement for the Use of Animals in Ophthalmic and Vision Research. Healthy male New Zealand white rabbits (Charles River, Wilmington, MA), weighing between 2.0 and 3.0 kg, were housed in separate cages. The rabbits were maintained in a controlled environment with a 12-hour on/off light cycle, and food and water were administered ad libitum. All procedures were performed with rabbits under anesthesia induced with an intramuscular injection of ketamine hydrochloride (35 mg/kg body weight) and xylazine hydrochloride (5 mg/kg body weight; both from Henry Schein, Dublin, OH).

Experimental Procedure

Celecoxib (LC Laboratories, Woburn, MA) was acquired in powder form, dissolved in dimethyl sulfoxide (DMSO; Sigma-Aldrich, St. Louis, MO), and processed through a 0.22-μm Millipore filter (Millipore Corporation, Bedford, MA). The following concentrations of celecoxib were prepared: 15, 30, and 60 mg/mL. The right eye (four eyes per concentration) of each animal was injected with 0.1 mL of a drug concentration, and the corresponding left eye (control) was injected with 0.1 mL DMSO.

Animals were anesthetized with xylazine hydrochloride and ketamine hydrochloride by intramuscular injection. Pupils were dilated with topical tropicamide 0.5% (Bausch & Lomb, Inc., Tampa, FL), phenylephrine 2.5%, and atropine sulfate 1% (both from Alcon, Fort Worth, TX). Proparacaine 0.5% (Alcon) was applied for corneal anesthesia. After application of 5% povidone-iodine solution (Alcon), prophylactic removal of 0.1 mL fluid from the anterior chamber was performed. Immediately afterward, a 25-gauge needle on a 1-mL tuberculin syringe was inserted 2 mm posterior to the limbus in the superotemporal quadrant. The needle was directed to the midvitreous, and the material was slowly injected. Intraocular pressure after the procedure was less than 30 in all eyes.

Examinations

Eyes underwent baseline slit-lamp biomicroscopy, indirect ophthalmoscopy, intraocular pressure (IOP) measurement, and electroretinographic (ERG) testing. Animals were checked daily, and complete ophthalmic exams including IOP measurements were repeated immediately after injection and at 1, 4, and 12 weeks. Electroretinographic testing was repeated at 1, 4, and 12 weeks postinjection.

Electrophysiology

Standard Ganzfeld scotopic (dark-adapted) and photopic (light-adapted) ERGs were obtained by masked trained personnel after anesthesia and pupillary dilation. Animals were dark adapted for at least 1 hour and were anesthetized 10 minutes before ERGs were obtained. Electroretinographic waveforms were obtained from each eye simultaneously by positioning the active electrodes (ERG-jet monopolar contact lens electrodes; Universo Plastique SA, Le Cret-Du-Locle, Switzerland) on each cornea. The reference and ground electrodes (Grass subdermal needle electrode; Astro-Med, Inc., West Warwick, RI) were placed subcutaneously in the nasal bridge and pinna, respectively.

Full-field dark- and light-adapted ERG recordings were obtained using the LKC UTAS-E3000 system (LKC Technologies, Inc., Gaithersburg, MD). The standard white flash illumination was provided by Ganzfeld 2503D stimulator (LKC Technologies, Inc.). The software LKC EM for Windows (EMwin v3.0; LKC Technologies, Inc.) was used to control the recording setting and analyze the data. Signal amplification, luminance calibration, and band pass filtering are integrated into the LKC system. The band pass filter was set between 0.3 and 500 Hz.

Three ERG recordings were obtained per stimulus intensity. The scotopic and photopic white flash stimuli were performed with an intrastimulus interval of 1 minute. The intrastimulus recovery interval allowed the retina to recover from the previous flash.

Scotopic ERG responses were evoked using flash intensities of –34, –24, –12, 0, 7.5, and 15 dB. Rabbits were subsequently light adapted under steady background adapting field of 30 cd/m² to saturate the rod photoreceptors. The photopic ERG responses were evoked with flash intensities of –2, 0, 2.5, 10, and 5 dB for a 30-Hz flicker.

ERG Analysis

Scotopic (dark-adapted) and photopic (light-adapted) amplitudes of the a-wave and b-wave were measured. A-wave amplitudes were measured from the preresponse baseline to the trough of the negative wave and reflect photoreceptor function (outer retina). B-wave amplitudes were measured from the trough of the a-wave to the peak of the b-wave and reflect Müller cell and bipolar cell function (inner retina). For oscillatory potentials of the 30-Hz flicker response, the amplitude was measured from the preresponse baseline to the peak.

Histology

Immediately following the 12-week ERG, animals were killed with intracardiac pentobarbital overdose (200 mg), and the eyes were enucleated and fixed in 10% formalin for 24 hours. Gross examination was performed. Representative sections of the optic nerve head, medullary ray, and inferior retina (region of dependency where the drug would likely settle) were paraffin embedded. Semithin sections were stained with hematoxylin–eosin and periodic acid–Schiff for light microscopy.

Rabbit Model of Endotoxin-Induced Uveitis (EIU)

A 1 mg/mL stock solution of endotoxin (lipopolysaccharide from Salmonella typhimurium; Sigma-Aldrich) was diluted immediately before injection into isotonic saline under sterile conditions and processed through a 0.22-μm Millipore filter (Millipore Corporation) to arrive at a final concentration of 1 μg/50 μL. Celecoxib was prepared in DMSO at a concentration of 60 mg/mL. Rabbits were anesthetized, and then proparacaine 0.5%, atropine 1% (to reduce discomfort), and povidone-iodine 5% were applied to the ocular surface before injection. A total of 12 rabbits were injected in both eyes with 1 μg endotoxin using a 25-gauge needle inserted 2 mm posterior to the limbus in the superotemporal quadrant. After 5 minutes, six right eyes of six rabbits were injected with 3 mg (50 μL) celecoxib, and six right eyes of six different rabbits were injected with 2 mg (50 μL) triamcinolone acetonide (40 mg/mL; Alcon). All left eyes served as controls and were injected with equal-volume saline.

Measurement of Intraocular Inflammation and Prostaglandin E₂

Twenty-four hours after injection of endotoxin, 200 μL aqueous was removed by anterior chamber paracentesis using...
a 25-gauge needle on a tuberculin syringe. Total leukocyte counts were immediately performed by a masked grader (RS) using a hemocytometer. The remaining sample was immediately diluted 1:1 in chilled assay buffer (Assay Designs, Ann Arbor, MI) and frozen immediately at −80°C. Animals were then euthanized. Prostaglandin E₂ (PGE₂) levels were later determined by enzyme-linked immunoassay according to the manufacturer’s instructions (Assay Designs).

**Pharmacokinetics**

For pharmacokinetic testing, both eyes of rabbits were injected with 3 mg celecoxib in the manner described above, and the animals were then euthanized at the following time points after injection: 0.25, 1, 4, 24, and 72 hours and 1, 2, 4, and 8 weeks. Eyes were enucleated and immediately snap frozen in liquid nitrogen for later isolation of the vitreous and retina/choroid. Drug concentrations in the vitreous and retina/choroid were measured by high-performance liquid chromatography (HPLC) and tandem mass spectrometry (two eyes per time point).

A QTrap4500 triple quadrupole mass spectrometer (Applied Biosystems, Foster City, CA) coupled with a Shimadzu liquid chromatography system (Shimadzu LC20-AD, Columbia, MD) was used for the study. Celecoxib concentrations in rabbit ocular tissue samples were measured by means of electrospray ionization tandem mass spectrometry (MS/MS) analysis after separation by HPLC. Analytes were separated on Zorbax C18 Column (4.6 × 50 mm, 5 μm; Agilent Technologies, Santa Clara, CA), using acetonitrile with 0.1% formic acid (organic phase) and 5 mM ammonium formate in water, adjusted to pH 3.5 with formic acid (aqueous phase) as mobile phase in gradient elution mode with flow rate of 1.0 mL/min. Total run time of analysis was 3.6 minutes per run. Celecoxib (LC Laboratories) and nimesulide (Sigma-Aldrich, internal standard) were analyzed in negative ionization mode using the following multiple reaction monitoring (MRM): 379.5/316.0 (celecoxib) and 306.9/228.60 (nimesulide).

Soluble and insoluble celecoxib present in vitreous humor was estimated using the following method. Vitreous humor was centrifuged at 10,000g (AccuSpin; Fisher Scientific, Pittsburgh, PA) for 5 minutes, and the vitreous humor free of any particulate was separated from the pellet. For estimating the soluble celecoxib, 0.125 mL vitreous humor free of particulate was added to tubes with 0.25 mL water containing 100 ng/mL internal standard (nimesulide). Tissues were vortexed for 15 minutes on a multiple vortexer (VX-2500, VWR; LabShop, Batavia, IL). Subsequently, 0.75 mL acetonitrile was added to the sample mixture, and the tubes were vortexed.
for 30 minutes. Sample preparations were centrifuged at 10,000g for 5 minutes to separate the tissue proteins. The supernatant was pipetted out and transferred to glass tubes, and the solvent was evaporated under nitrogen stream (Multi-Evap; Organomotion, Berlin, MA) at 40°C. The residue after evaporation was reconstituted with 0.125 or 0.25 mL acetonitrile-water (50:50 vol/vol) and diluted as needed before injecting onto the LC-MS/MS.

For estimating the insoluble celecoxib in vitreous humor, pellet obtained from the centrifugation of vitreous humor was added with 1.0 mL acetonitrile containing 10 μg/mL internal standard. Subsequently, samples were vortexed for 10 minutes to dissolve all celecoxib. Further, the resulting homogenate was diluted 1000 times with acetonitrile-water mixture (1:1) before injecting onto the LC-MS/MS.

In the case of retina/choroid both soluble and insoluble celecoxib were measured together using the following procedure. Twenty milligrams of retina/choroid was weighted into Eppendorf tubes and added with 0.25 mL water containing 100 ng/mL internal standard. Samples were homogenized using a handheld homogenizer (Tissue-Tearor; Biospec Products, Bartlesville, OK) on an ice bath for 15 to 30 seconds such that the tissue was completely homogenized. Subsequently, 0.75 mL acetonitrile was added to the sample mixture, and the tubes were vortexed for 30 minutes to extract the drug into acetonitrile. Sample tubes were centrifuged at 10,000g for 5 minutes; the supernatant was transferred to glass tubes, and the solvent was evaporated under nitrogen at 40°C. The residue after evaporation was reconstituted with 0.125 or 0.25 mL acetonitrile-water (50:50 vol/vol) and further diluted as needed before injecting onto the LC-MS/MS.

**Data Analysis**

All data are reported as mean with standard deviation unless otherwise noted. Statistical significance was determined using a two-tailed Student's *t*-test. *P* values < 0.05 were considered significant.

Figure 2. Intraocular pressure measurements taken at baseline and 1, 4, and 12 weeks. There were no significant differences among increasing doses of celecoxib when compared to control left eyes injected with vehicle (DMSO).

Figure 3. Representative photographs taken before injection (left) and immediately after injection (right) of 3 mg celecoxib. Precipitated drug is clearly apparent behind the lens.
RESULTS

Clinical Observations

All animals tolerated the injections well with no signs of pain or inflammation. Examination at baseline and 1, 4, and 12 weeks after injection demonstrated no intraocular inflammation, but some eyes injected with celecoxib had cataract formation that was dose dependent (Fig. 1). At 12 weeks, one of four eyes injected with 1.5 mg celecoxib showed a mild cataract; two of four eyes injected with 3 mg had mature white cataracts; and all four eyes injected with 6 mg had mature cataracts. In contrast, only 1 of 12 control eyes had a central cataract (presumed to be due to trauma). No differences in IOP between celecoxib and control eyes were observed at any time point (Fig. 2). Indirect ophthalmoscopy in eyes without cataract formation demonstrated no signs of hemorrhage, whitening, or optic nerve pallor. In eyes injected with celecoxib, white drug precipitate was observed behind the lens immediately after injection (Fig. 3), which disappeared completely by 12 weeks. There were no cases of retinal detachment or endophthalmitis.

ERG Waveform

Representative baseline and 1-, 4-, and 12-week scotopic and photopic ERG waveforms are shown in Figures 4 and 5, respectively. All eyes injected with 1.5, 3, or 6 mg celecoxib showed similar waveform patterns with clearly distinguishable a-waves (negative wave) and b-waves (positive wave) when compared to control eyes injected with vehicle (DMSO).

ERG Amplitudes

Mean photopic a-wave and b-wave amplitudes are shown in Table 1. There were no significant differences between celecoxib-injected right eyes and DMSO-injected left eyes at any time point. Mean scotopic a-wave and b-wave amplitudes are shown in Table 2. There was a decrease in a-wave amplitude at 1 week in eyes injected with 3 mg celecoxib, but this decrease was not observed at subsequent time points or at other doses of celecoxib. There were no differences in b-wave amplitudes between celecoxib-injected right eyes and DMSO-injected left eyes at any time point.

Histology

Light microscopy demonstrated absence of intraocular inflammation in sectioned eyes. Cataract formation was clearly present in eyes injected with celecoxib, but no signs of focal or diffuse inner and outer retinal atrophy or necrosis were observed (Fig. 6). There were no signs of optic nerve toxicity in any of the sections examined (Fig. 6). There was mild
vacuolization in the ganglion cell layer and disruption of photoreceptor outer segments in both treated and control eyes, to the same degree, consistent with autolysis and tissue processing artifacts.

**Pharmacokinetics**

Measured concentration in the vitreous humor and retina/choroid at specific time points after a single injection of 3 mg celecoxib is presented in Table 3. There were detectable levels of drug in the retina/choroid at 8 weeks that exceeded the median inhibitory concentration (15 ng/mL)\textsuperscript{13} of celecoxib for COX-2. Drug precipitate was isolated at each time point and measured separately.

**Efficacy in Endotoxin-Induced Model of Uveitis**

Injection of endotoxin in the rabbit vitreous induced a robust inflammatory response at 24 hours, reflected in a mean aqueous leukocyte count of 13,400 ± 7052 cells/µL (Fig. 7).

**Table 1.** Mean Photoptic A-Wave and B-Wave Amplitudes

<table>
<thead>
<tr>
<th></th>
<th>Baseline</th>
<th>Week 1</th>
<th>Week 4</th>
<th>Week 12</th>
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<td></td>
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<td>R</td>
<td>L</td>
<td>P</td>
<td>R</td>
<td>L</td>
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<tr>
<td>Mean a-wave amplitude (SD) for animals treated with celebrex in the right eye and DMSO in the left eye</td>
<td></td>
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<tr>
<td>All groups</td>
<td>−16 (2)</td>
<td>−15 (2)</td>
<td>0.9034</td>
<td></td>
</tr>
<tr>
<td>1.5 mg</td>
<td>−19 (6)</td>
<td>−27 (12)</td>
<td>0.539</td>
<td>−23 (6)</td>
</tr>
<tr>
<td>3.0 mg</td>
<td>−17 (3)</td>
<td>−17 (2)</td>
<td>0.980</td>
<td>−14 (2)</td>
</tr>
<tr>
<td>6.0 mg</td>
<td>−22 (11)</td>
<td>−27 (14)</td>
<td>0.781</td>
<td>−34 (21)</td>
</tr>
</tbody>
</table>

Mean b-wave amplitude (SD) for animals treated with celebrex in the right eye and DMSO in the left eye

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<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>All groups</td>
<td>147 (10)</td>
<td>136 (13)</td>
<td>0.255</td>
<td></td>
</tr>
<tr>
<td>1.5 mg</td>
<td>138 (27)</td>
<td>145 (24)</td>
<td>0.850</td>
<td>140 (15)</td>
</tr>
<tr>
<td>3.0 mg</td>
<td>118 (23)</td>
<td>140 (8)</td>
<td>0.407</td>
<td>165 (40)</td>
</tr>
<tr>
<td>6.0 mg</td>
<td>114 (12)</td>
<td>116 (5)</td>
<td>0.873</td>
<td>216 (42)</td>
</tr>
</tbody>
</table>

**Figure 5.** Representative photopic waveforms measured at baseline and 1, 4, and 12 weeks using light intensity of 2.5 dB. The celecoxib waveform was averaged from the four right eyes injected with 3 mg celecoxib, and the vehicle (control) waveform was averaged from the four corresponding left eyes injected with DMSO.
Concurrent treatment with 3 mg celecoxib and 2 mg triamcinolone markedly decreased the inflammatory response to $8094 \pm 6400$ cells/µL ($P = 0.02$) and $9222 \pm 5100$ cells/µL ($P = 0.01$), respectively. Overall, celecoxib and triamcinolone reduced inflammation by 40% and 31%, respectively. Aqueous PGE2 levels rose to $7140 \pm 7020$ pg/mL 24 hours after injection of endotoxin (Fig. 8). Prostaglandin E2 levels were reduced by 61% to $2769 \pm 2344$ pg/mL ($P = 0.04$) and 83% to $1209 \pm 1057$ pg/mL ($P < 0.01$) in celecoxib- and triamcinolone-treated eyes, respectively.

**DISCUSSION**

The results of this study demonstrate that a single intraocular injection of celecoxib appears to be nontoxic to the retina and provides sustained drug delivery out to 8 weeks. Our results...
also demonstrate that intraocular celecoxib significantly reduces intraocular inflammation in an animal model of uveitis. To our knowledge, we are the first to report these findings, which may have considerable importance since celecoxib does not increase IOP and thereby offers a distinct clinical advantage over corticosteroids.

Inflammation is mediated in part by the COX pathway, which leads to the production of PGs. Five classes of PGs exist (PGE2, prostaglandin D2, prostaglandin F2α, prostaglandin I2, thromboxane A2), which are biosynthesized from membrane-bound arachidonic acid by COX enzyme. Two isoforms, COX-1 and COX-2, are firmly established and are among the most thoroughly studied and best-understood mammalian enzymes. In the human retina, COX enzyme can be detected in retinal endothelial cells, astrocytes, microglia, ganglion cells, amacrine cells, Müller cells, and retinal pigment epithelium cells and is upregulated in response to inflammatory cytokines. Within the eye, PGs promote vasodilation, disrupt the blood–ocular barrier, facilitate leukocyte migration, and interact with and amplify other soluble mediators including VEGF. Consequently, their inhibition has favorable effects on inflammation and ME.

Inflammation contributes to DR and AMD, and several lines of evidence indicate that PGs play an important role. Cyclooxygenase-2 enzymes have been detected in choroidal neovascular membranes, and pharmacologic inhibition or genetic deletion of COX reduces choroidal neovascularization. In animal models of DR, PGs induce VEGF production with subsequent vascular leakage and retinal neovascularization. Prostaglandin E2 is increased by 40% in the retinal vasculature of diabetic rats, and NSAIDs significantly inhibit diabetes-induced retinal microvascular disease and vascular leakage in animal models. In support of experimental evidence, inflammatory cytokines are consistently elevated in the vitreous of patients with advanced stages of DR, and prospective studies have reported favorable effects of NSAID treatment on vascular leakage and progression of retinopathy.

Despite increasing rationale to test the efficacy of NSAIDs to treat AMD and DR, there is no convincing evidence that topical or systemic NSAIDs measurably reduce retinal PG levels. In contrast, intraocular administration results in considerably higher retinal drug levels while minimizing systemic exposure. Previous studies have demonstrated that intravitreal injection of ketorolac results in therapeutic retinal drug levels 100 to 1000 times greater than after topical application. However, ketorolac is six times more selective for COX-1 than COX-2 and has a short intraocular half-life, which greatly limits its therapeutic effect.

Celecoxib, on the other hand, exhibits a prolonged intraocular half-life due to its relative insolubility in vitreous. After a single intravitreal injection, the majority of drug rapidly precipitates (similarly to triamcinolone acetonide) but still maintains a solubilized concentration far exceeding its median

### Table 3. Intraocular Pharmacokinetics of Celecoxib

<table>
<thead>
<tr>
<th>Time Point</th>
<th>Vitreous Humor, µg/mL</th>
<th>Retina/Choroid, µg/g</th>
<th>Insoluble Celecoxib, µg</th>
</tr>
</thead>
<tbody>
<tr>
<td>15 min</td>
<td>128.12</td>
<td>800.62</td>
<td>426.26</td>
</tr>
<tr>
<td>1 h</td>
<td>121.14</td>
<td>562.67</td>
<td>187.47</td>
</tr>
<tr>
<td>4 h</td>
<td>72.63</td>
<td>82.48</td>
<td>320.71</td>
</tr>
<tr>
<td>24 h</td>
<td>164.75</td>
<td>420.22</td>
<td>241.43</td>
</tr>
<tr>
<td>72 h</td>
<td>13.31</td>
<td>15.50</td>
<td>45.71</td>
</tr>
<tr>
<td>1 wk</td>
<td>14.33</td>
<td>227.74</td>
<td>344.14</td>
</tr>
<tr>
<td>2 wk</td>
<td>16.13</td>
<td>16.81</td>
<td>347.69</td>
</tr>
<tr>
<td>4 wk</td>
<td>0.15</td>
<td>8.56</td>
<td>276.22</td>
</tr>
<tr>
<td>8 wk</td>
<td>0.06</td>
<td>132.31</td>
<td>264.62</td>
</tr>
</tbody>
</table>

Median inhibitory concentration of celecoxib for COX-2 is 0.015 µg/mL. Data represented as mean values for n = 2 eyes. Vitreous humor concentrations refer to only soluble celecoxib. Insoluble celecoxib refers to the celecoxib precipitate present in the isolated vitreous humor (range, 0.82–1.37 mL). Concentrations in the retina/choroid represent both soluble and insoluble celecoxib.

![Anterior Chamber Inflammation](https://tvst.arvojournals.org/)

**Figure 7.** Anterior chamber inflammation 24 hours after endotoxin administration. There is a significant reduction in inflammatory cells in eyes injected with 3 mg celecoxib (P = 0.02) and 2 mg triamcinolone (P = 0.01) when compared to control eyes injected with saline.
inhibitory concentration for COX-2. Celecoxib is also 300 times more selective for COX-2 than for COX-1. Cyclooxygenase-2 is the inducible isoform that is upregulated in the presence of proinflammatory cytokines in retinal cells. In contrast, COX-1 contributes to normal physiological processes and is expressed in a wide variety of tissue under normal circumstances. Consequently, sustained inhibition of COX-1 may not be desirable and may result in toxicity. In addition, although all NSAIDs inhibit COX enzyme, celecoxib also inhibits lipoxygenase (LPO) enzyme, which leads to the production of proinflammatory leukotrienes. This unique quality of celecoxib may bestow upon it greater anti-inflammatory properties than other NSAIDs (corticosteroids inhibit both COX and LPO enzymes). For these reasons, celecoxib offers ideal characteristics for sustained intraocular anti-inflammatory effects.

There was no observed increase in IOP after intraocular injection of celecoxib, but we were surprised by the high degree of cataract formation. To our knowledge this side effect has not been reported previously with systemic use, and to date there has been no consistent association between NSAIDs (corticosteroids inhibit both COX and LPO enzymes). For these reasons, celecoxib offers ideal characteristics for sustained intraocular anti-inflammatory effects.

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In conclusion, our data show that intravitreal celecoxib appears to be nontoxic in rabbit eyes. Despite the possibility of cataract formation, there was no increase in IOP, and the drug was well tolerated with no other evident adverse effects. Since the vitreous volume of adult eyes is much greater than in rabbits, doses greater than 6 mg may be safe. Intraocular celecoxib significantly reduced inflammation and PG production and achieved sustained therapeutic drug levels in the retina out to 8 weeks after a single injection. These results demonstrate the potential feasibility of intraocular celecoxib to treat ME, DR, and AMD, particularly in patients at risk for glaucoma; but further studies are needed to confirm and expand upon these initial findings.

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