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Brief Report

Lens Concentration of Ofloxacin and Lomefloxacin in an Experimental Endophthalmitis Model

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ABSTRACT

Background: Bacterial endophthalmitis is a serious complication of ocular surgery and penetrating trauma. The primary causative organisms are strains of *Staphylococcus aureus* and *Staphylococcus epidermidis*. Fluoroquinolones are widely used to treat endophthalmitis. There are a few studies on the penetration of fluoroquinolones into the lens in inflamed eyes. A literature search did not identify any data regarding penetration of topical ofloxacin into the lens in normal and inflamed eyes.

Objective: The aim of this study was to determine the penetration of topical ofloxacin and lomefloxacin into the lens in a rabbit endophthalmitis model.

Methods: New Zealand white rabbits were randomly divided into 2 groups. The left eyes were infected with an intravitreal inoculation of *S aureus*. The right eyes were used as a noninoculated control. Groups 1 and 2 received topical ofloxacin and lomefloxacin treatment, respectively, 24 hours after the inoculation. Two drops of the study drugs were instilled in the eyes every 30 minutes for 3 hours and then every 60 minutes for 3 hours. Lens samples were obtained 30 minutes after the last ofloxacin or lomefloxacin drops were administered. High-performance liquid chromatography was used to determine the fluoroquinolone concentration.

Results: Ten rabbits were equally divided into the 2 treatment groups. There was no significant difference in mean (SD) lens concentrations between the control and inoculated eyes in either treatment group—ofloxacin (0.26 [0.32] μg/mL vs 0.11 [0.05] μg/mL, respectively) and lomefloxacin (0.50 [0.87] μg/mL vs 0.12 [0.08] μg/mL, respectively).

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Conclusions: The results of this small experimental study found that topical ofloxacin and lomefloxacin can accumulate in the crystalline lens after installation. Inflammation did not affect the penetration of ofloxacin or lomefloxacin into the lens. (*Curr Ther Res Clin Exp.* 2007;68:184–190)

Key words: experimental endophthalmitis, fluoroquinolone, ofloxacin, lomefloxacin, lens, animal study, rabbit.

INTRODUCTION

The first quinolone, nalidixic acid, was introduced in 1962.[1] Since then, structural modifications have been made and quinolones have improved gram-positive coverage. Quinolones block bacterial DNA synthesis by inhibiting topoisomerase enzymes. Quinolones are bactericidal and are active against a variety of gram-negative and gram-positive bacteria.[2,3]

Although there have been several studies[4–10] of the penetration of topical fluoroquinolones into the aqueous and vitreous humor, reports on penetration of these antimicrobials into the lens are limited.[11,12] Furthermore, a literature search (MEDLINE, years 1980–2006, English language; key terms: *fluoroquinolone*, *ofloxacin*, *lomefloxacin*, and *lens penetration*) did not identify any data on lens penetration of topically administered ofloxacin in normal and inflamed eyes. It is important to determine the amount of drug that penetrates into the lens because the lens may act as a reservoir of active agents, meaning that excessive use may result in accumulation of the drug and may cause opacities.

The aim of this study was to determine and compare the concentrations of ofloxacin and lomefloxacin in the lens after topical instillation in normal and inflamed eyes.

METHODS

New Zealand white rabbits weighing between 2.1 and 3.0 kg were equally divided into 2 groups (groups 1 and 2). The animals were treated according to the Association for Research in Vision and Ophthalmology Statement for the Use of Animals in Ophthalmic and Vision Research.[13] Each animal was housed in a separate cage and was given food and water ad libitum. The study protocol was reviewed and approved by the Ankara Training and Research Hospital ethics committee, Ankara, Turkey.

Endophthalmitis Model

All rabbits were anesthetized using an IM injection of xylazine hydrochloride 10 mg/kg and ketamine hydrochloride 50 mg/kg. All intravitreal injections were made ~2 mm from the superonasal limbus with a 27-gauge needle attached to a 1-mL tuberculin syringe (TIBSET A.S., Istanbul, Turkey). The left eye of each animal was used for study drug injections, whereas the right eye was used as a

noninoculated control. *Staphylococcus aureus* (ATCC® 29213, American Type Culture Collection, Manassas, Virginia) was used to induce intraocular infection.

Fluoroquinolone Treatment

Twenty-four hours after bacterial inoculation, 2 drops of commercially available ofloxacin 0.3% (Exocin®, Allergan, Westport, Ireland) and lomefloxacin 0.3% (Okacin®, Novartis Pharma AG, Basel, Switzerland) were instilled into the eyes of the animals in group 1 and 2, respectively, every 30 minutes for 3 hours and then every 60 minutes for 3 hours. Thirty minutes after the last fluoroquinolone administration, lenses were extracted and samples were stored at –80°C until analysis.

The investigators who instilled the drops were not masked to the study group, while the investigators who analyzed the lens samples were masked.

High-Performance Liquid Chromatography of Fluoroquinolone Concentrations

Concentrations of ofloxacin and lomefloxacin in the lens samples were measured using high-performance liquid chromatography, as described previously.[14,15] The separation was performed on a Radial-pak, C18 cartridge column (100 × 8 mm i.d., particle size 10 mm) and precolumn (C18) (Waters Corp., Milford, Massachusetts). The sample elution was monitored on a fluorescence detector using excitation and emission wavelengths of 280 and 455 nm, respectively. The mobile phase consisted of acetonitrile and 0.1 M of sodium dihydrogen phosphate (2:8 [v/v]; pH = 3.9). The mobile phase was delivered at a flow rate of 2 mL/min. Quinine sulfate was used as an internal standard (2 mg/mL). Lens samples were homogenated and centrifuged at 8000 rpm for 5 minutes. The supernatant was directly injected after dilution and addition of quinine sulfate; 20 µL of aliquot were injected through the column. The within-day and day-to-day precision values were <9% for the 2 drugs at 0.05, 0.1, and 0.4 mg/mL (n = 6); the within-day and day-to-day accuracy values were in the range of 96.3% to 102.7% for the drugs at the concentrations given; the detection limit corresponding to a signal-to-noise ratio of 3:1 was 0.6 ng/mL.[14,15]

Statistical Analysis

Data were expressed as mean (SD) and range. Concentrations of fluoroquinolones in normal and infected lenses were compared using the Kruskal-Wallis test, a nonparametric test for paired samples. $P < 0.05$ was considered statistically significant. All statistical analyses were carried out using Statistical Packages for Social Sciences (SPSS) software version 10.0 (SPSS Inc., Chicago, Illinois).

RESULTS

The mean (SD) lens concentrations of the control and inoculated eyes for ofloxacin (0.26 [0.32] µg/mL and 0.11 [0.05] µg/mL, respectively) and lomefloxacin (0.50 [0.87] µg/mL and 0.12 [0.08] µg/mL, respectively) are shown in

Tables I and **II**, respectively. There were no significant differences between the 2 groups.

DISCUSSION

Based on a search of the literature, this is the first study reporting topical ofloxacin drug concentrations in the crystalline lens. In this study, inflammation did not affect lens penetration of topically applied ofloxacin and lomefloxacin. These results suggest the need for more studies of ofloxacin in the treatment of ocular lens infectious diseases, particularly in lenticular abscesses.

Superficial and deep ocular infections, such as conjunctivitis, corneal ulcers, and endophthalmitis, are caused by a diverse group of bacterial, viral, and fungal pathogens. Bacterial endophthalmitis most commonly occurs as a postoperative complication, but it may also result from penetrating ocular trauma or systemic

Table I. Lens concentrations of ofloxacin in the normal (control) and *Staphylococcus aureus*–inoculated eyes of New Zealand white rabbits (n = 5).

	Topical Ofloxacin Concentration, μg/mL	
Rabbit	Control Eye	Inoculated Eye
1	0.07	0.12
2	0.08	0.06
3	0.13	0.18
4	0.19	0.13
5	0.83	0.06
Mean (SD)*	0.26 (0.32)	0.11 (0.05)

*No statistically significant difference was observed.

Table II. Lens concentrations of lomefloxacin in the normal (control) and *Staphylococcus aureus*–inoculated eyes of New Zealand white rabbits (n = 5).

	Topical Lomefloxacin Concentration, μg/mL	
Rabbit	Control Eye	Inoculated Eye
1	2.05	0.23
2	0.12	0.08
3	0.05	0.16
4	0.06	0.07
5	0.22	0.04
Mean (SD)*	0.50 (0.87)	0.12 (0.08)

*No statistically significant difference was observed.

infection.[16,17] Despite the best available treatment, the treatment success in the patients with endophthalmitis is still insufficient. *Staphylococcus epidermidis* and *S aureus* are primary causes of postoperative bacterial endophthalmitis.[18] Topical fluoroquinolones are used in the treatment of ocular infectious diseases.[19]

The penetration of topical fluoroquinolones into several ocular tissues has been well documented.[5,7,11,20] Topical ofloxacin has been reported in clinical studies to penetrate ocular structures significantly better than the other first- and second-generation fluoroquinolones.[21,22] In the present study, we found the mean lens drug concentration to be 0.26 μg/mL (range, 0.07–0.83 μg/mL) for ofloxacin and 0.50 μg/mL (range, 0.05–2.05 μg/mL) for lomefloxacin in control eyes.

Aqueous and vitreous humor penetration of ofloxacin has been shown to increase in the inflamed eye.[23,24] In the current study, topical ofloxacin and lomefloxacin yielded a mean concentration of 0.11 μg/mL (range, 0.06–0.18 μg/mL) and 0.12 μg/ml (range, 0.04–0.23 μg/mL) in the lens of inoculated eyes, respectively. Inflammation did not significantly affect lens concentrations of either ofloxacin or lomefloxacin compared with the control eyes. In addition, these concentrations were lower than the MIC of *S aureus*, which is 0.63 μg/mL for ofloxacin[25] and 0.78 μg/mL for lomefloxacin.[26] Although the dosing regimen we used in this study did not reach a sufficient concentration to cover the infecting pathogens, different dosing regimens and different application methods (ie, oral plus topical) might allow the drugs to reach therapeutic concentrations.

This study supports the lenticular presence of topical ofloxacin and lomefloxacin. The small number of rabbits used was a limitation for the comparison of the drug concentrations between normal and inflamed eyes. For example, assuming a power of 90% and an α of 0.05, ~20 rabbits per group would be needed to show a significant difference. Another limitation of the study was the instillation method, which did not allow precise control of the amount of antibiotic instilled.

CONCLUSIONS

These results indicate that topical ofloxacin and lomefloxacin can accumulate in the lens and inflammation does not influence lens penetration of these 2 fluoroquinolones. Further studies are needed to demonstrate the effect of treatment protocol on the concentrations of topical fluoroquinolones in the lens and to investigate the effect of fluoroquinolone accumulation in the human lens.

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