CENTER FOR DRUG EVALUATION AND RESEARCH

APPLICATION NUMBER:

21-571

PHARMACOLOGY REVIEW

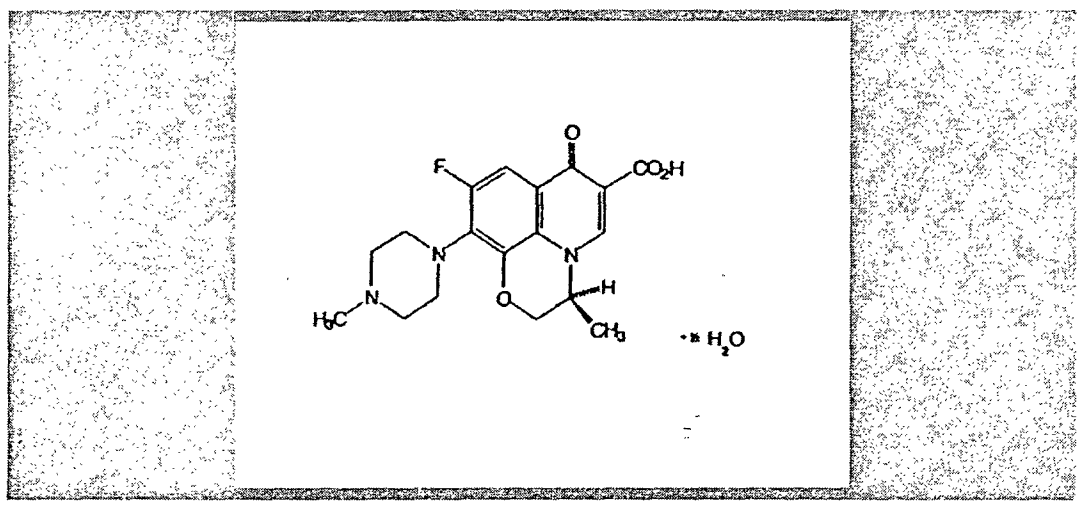
PHARMACOLOGY/TOXICOLOGY COVER SHEET

NDA number: 21-571
Review number: One
Sequence number/date/type of submission: 000, April 30, 2003, 505(b)1
Information to sponsor: Yes ( ) No (X)
Sponsor and/or agent: Santen Incorporated, 555 Gateway Drive, Napa, CA 94558
Manufacturer for drug substance: Daiichi Pharmaceutical Company, Tokyo 104, Japan.

Reviewer name: Asoke Mukherjee, Ph.D.
Division name: Anti-inflammatory, Analgesic and Ophthalmic Drug Products
HFD #: 550
Review completion date: Aug 18, 2003

Drug:

Trade name: Iquix® (Proposed)
Generic name (list alphabetically): 1.5% Levofloxacin ophthalmic solution
Code name: DR-3355
Chemical name: (S)-9-fluoro-2,3-dihydro-3-3-methyl-10- (4-methyl-1-piperazinyl)-7-oxo-7H-pyrido[1,2,3-de]-1,4-benzoxazine-6-carboxylic acid hemihydrate.
CAS registry number: 138199-71-0
Mole file number:
Molecular formula/molecular weight: $C_{18}H_{20}FN_3O_4,1/2H_20$; 370.38
Structure:



Relevant INDs/NDAs/DMFs: NDA 21-199, NDA 20-634, NDA 20-635, DMF —— IND 36,627, IND 38,368 and IND —

Reviewer Name: Asoke Mukherjee, Ph.D. NDA 21-571

Drug class: Antimicrobial agent

Indication: Corneal Ulcer

Clinical formulation:

Ingredient	Percent (w/v)	mg/ml
Levofloxacin	1.50	15.0
Glycerin, USP	—	—
HCl/NaOH	To pH 6.5	
Purified water	—	

Route of administration: Ophthalmic drops

Proposed use: Treatment of corneal ulcer caused by susceptible strains of bacteria

Disclaimer: Tabular and graphical information is from sponsor's submission unless stated otherwise.

APPEARS THIS WAY
ON ORIGINAL

Reviewer Name: Asoke Mukherjee, Ph.D. NDA 21-571

Executive Summary

I. Recommendations

A. Recommendation on Approvability: Levofloxacin 1.5% ophthalmic solution is recommended for approval from the non-clinical safety point of view with minor modifications of labeling as addressed below.

B. Recommendation for Nonclinical Studies: Nil

C. Recommendations on Labeling: Page M1-vol 01, 052 provided a proposed label of the product. The carcinogenicity, mutagenicity, impairment of fertility, and pregnancy information of 1.5% levofloxacin is similar to that already approved for NDA 21-199 (0.5% levofloxacin). However, animal to human dose ratios needs to be modified based on the recommended dose of the product.

According to the proposed uses

For carcinogenicity, mutagenicity, impairment of fertility:

In a long-term carcinogenicity study in rats, levofloxacin exhibited no carcinogenic potential following daily dietary administration for 2 years; the highest dose (100 mg/kg/day) was **100** times the highest recommended human ophthalmic dose.

The mutagenicity portion of the label should be same as proposed.

Levofloxacin caused no impairment of fertility or reproduction in rats at oral doses as high as 360 mg/kg corresponding to **400** times the highest recommended human ophthalmic dose.

Pregnancy:

Teratogenic effects. Pregnancy category C:

Levofloxacin at oral doses of 810 mg/kg/day in rats, which corresponds to approximately **1000** times the highest recommended human ophthalmic dose, caused decreased fetal body weight and increased fetal mortality.

No teratogenic effect was observed when rabbits were dosed orally as high as 50 mg/kg/day, which corresponds to approximately **60** times the highest recommended maximum human ophthalmic dose, or when dosed intravenously as high as 25 mg/kg/day, corresponding to approximately **30** times the highest recommended human ophthalmic dose.

II. Summary of Nonclinical Findings

A. Brief Overview of Nonclinical Findings: Levofloxacin at 1.5% ophthalmic solution did not show any ocular and systemic toxicity in rabbits. However, 3% ophthalmic solution showed a reduction in the wound healing in the monkey model of experimental corneal ulcer. Based on the data, it is suggested that patients treated with 1.5% levofloxacin ophthalmic solution be monitored for healing of corneal ulcer following the completion of treatment.

B. Pharmacologic Activity:

Levofloxacin is a fluoroquinolone antibacterial agent, structurally related to nalidixic acid. The mechanism of antibacterial effect is considered to be due to the inhibition of DNA topoisomerase and gyrase that are implicated for the DNA replication and cell division. Pharmacodynamic activity of levofloxacin was demonstrated in normal and ulcerative keratitis models in rabbit eyes. The data showed that levofloxacin was more effective against gram negative Pseudomonas strains than gram positive Staphylococcus strains. However, data from an n-heptanol induced keratitis model in the monkey showed that levofloxacin delayed the corneal wound healing at 3.0% ophthalmic solution. The 1.5% solution did not show any deleterious effect on the corneal wound healing

C. Nonclinical Safety Issues Relevant to Clinical Use:

A repeated dose toxicity study was conducted in Dutch belted rabbits at 1.5 and 3.0% preservative free ophthalmic solutions for 28 days. Data did not show any ocular and systemic toxicity related to the levofloxacin treatment. A similar observation was reported for a 26-week ophthalmic safety study in rabbits and was reviewed under NDA 21-199.

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Reviewer Name: Asoke Mukherjee, Ph.D. NDA No.: 21-571

PHARMACOLOGY/TOXICOLOGY REVIEW

I. PHARMACOLOGY:

Primary pharmacodynamics:

Page 1, vol 1, M4 (PC016-16R).

The antibacterial effect of levofloxacin against Pseudomonas aeruginosa and Staphylococcus aureus were investigated. These two organisms represented common bacterial infections that lead to corneal ulcer. Levofloxacin (LVX) at 40, 15 and 5 μg/ml showed 99.9% lethality of the P. aeruginosa at 1, 2 and 4 hour incubation period. Similar concentrations of levofloxacin showed 99.9% lethality of S. aureus at 4 and 8 hours.

Above data suggest that levofloxacin is more effective against Pseudomonas than S. aureus infections. Lvofloxacin inhibited the growth of both organisms and inhibited the number of viable bacterial colonies in vitro. In another post antibiotic effect study (# PC016-15R, page 11, Vol 1, M4), the sponsor stated that exposure to high concentrations of the antibiotic with short exposure time increased the post antibiotic effect (PAE) of levofloxacin treatment against a gram negative Pseudomonas aeruginosa and gram positive Staphylococcus aureus in vitro. The PAE was the time taken for the organisms to grow following pretreatment with antibiotics compared to the untreated control. The longer PAE means the longer period it takes to grow after the contact time ended. The longer PAE means the less frequent dosing in the clinical situation. The sponsor also stated that the greater magnitude of PAE, the likelihood of resumption of bacterial growth between doses would be minimal. The PAE data for levofloxacin against the growth of P. aeruginosa and S. aureus are shown in the table below.

LVX (μg/ml)	Organism	Exposure (min)	PAE (hr)
40	P.aeruginosa	5	3.7
20	P.aeruginosa	20, 45	1.6, 2.1
15	S.aureus	5, 30	10.7, 10.4
5	P.aeruginosa	120	1.1
5	S.aureus	120, 240	1.5, 0.6

The high and low doses represent 5x and 1x of minimal inhibitory concentration (MIC_{90})

Mechanism of action: The mechanism of action of the drug was discussed in the review for NDA 21-199. Levofloxacin is the levo isomer of fluoroquinolone antibacterial agent, ofloxacin. Its mode of action is possibly inhibition of bacterial replication by the inhibition of transcription of DNA. It is also known to be an inhibitor of DNA topoisomerase and gyrase. Levofloxacin inhibits both gram positive and gram negative bacteria and is an effective broad spectrum antibacterial agent.

Drug activity related to proposed indication:

A comparative efficacy study with 1.5% levofloxacin versus 0.3% ofloxacin in treating an experimental model of Pseudomonas keratitis in female rabbits. Page 22, vol 1, M4, Study # PC016-04R.

The efficacy of 1.5% levofloxacin ophthalmic solution was investigated in Pseudomonas induced keratitis in the presence of experimental ulcer induced by heptanol in rabbits. About 10^6 colony-forming units (CFU) of P. aeruginosa was instilled into the right eye. Keratitis was induced within 24 hours. The left eye was considered to be the untreated control. Levofloxacin 1.5%, ofloxacin 0.3% or levofloxacin placebo was administered topically into the right eye at 24 hours after the inoculation of P. aeruginosa. The dose volume was 50 µL per dose. The sponsor stated that a new bottle of the test article was used each day. Eye drops were administered for two days. Quality assurance test for levofloxacin showed a concentration of [redacted] mg/ml levofloxacin in each vial. The study design is shown in the table below.

Group	N	Drug	Conc, mg/ml	Treatment Day 1, 50 µL/dose	Treatment Day 2, 50 µL/dose
1	10	Vehicle control	vehicle	10 doses, treatment every 2 hours up to 8 doses and 2 more doses within next 6 hour.	8 doses, treatment every 2 hours up to 8 doses
2	10	Levofloxacin	15 mg/ml	10 doses, treatment every 2 hours up to 8 doses and 2 more doses within next 6 hours.	8 doses. treatment every 2 hours up to 8 doses
3	10	Ofloxacin	3 mg/ml	31 doses, Treatment every 30 minutes up to 29 doses and two additional doses within next 6 hours.	29 doses, treatment every 30 minutes up to 29 doses.

Clinical examinations other than ocular changes were conducted once daily. Degree of infection was evaluated macroscopically and scored. Animals were euthanized at the end of the treatment, cornea was collected for quantitative bacterial analyses.

No mortality and clinical signs were reported due to the treatment. Keratitis and clinical infection were noted in all animals on day 2 except 3 animals from group 3. Clinical signs were partial to closed eyes, slight to moderate swelling of the eyelids, redness of the eye, cloudy

appearance and ocular discharge. The sponsor stated that clinical signs of the eye persisted until the end of the study.

Quantitative CFU showed no evidence of Pseudomonas in the cornea in groups 2 and 3 animals at the end of the treatment. The control animals showed CFU between 2-118x10^3 in nine out of 10 rabbits. Another control rabbit showed 2106x10^3 CFU.

Data show that levofloxacin, 1.5% ophthalmic drops at 18 drops over 48 hours, was effective in inhibiting bacterial growth in the rabbit eye after inducing ulcer and corneal epithelial damage by n-heptanol. The experiment was conducted at

Secondary pharmacodynamics:

Evaluation of levofloxacin ophthalmic solution on epithelial wound healing in heptanol-induced corneal ulcer in male cynomolgus monkeys. Study # PC016-03R, page 3434, vol 11, M 4.

The experiment was conducted in 10 male cynomolgus monkeys with 15 and 30 mg/ml ophthalmic solutions of levofloxacin. Each group had 5 monkeys. The test article was administered 4 times daily for 5 days into the right eye at 2 drops/eye. The left eye was untreated control. Prior to the treatment with levofloxacin, experimental corneal ulcer was induced in both eyes with heptanol. All animals were observed twice daily for clinical signs. Mortality was checked once daily. All animals were graded starting day 2 post dosing using a 0-3 scale. Both eyes were photographed before the treatment and daily up to day 11. The sponsor has not indicated whether day 1 was the first day of starting treatment with levofloxacin following n-heptanol induced corneal ulceration. The corneal thickness was measured at pretreatment and daily up to day 11. The body weight was recorded at pretreatment, day 1 and once a week. Slit lamp and indirect ophthalmoscopic examinations of the eye were performed on all animals once before the treatment and on day 8 after dilating the pupil with 1% Atropisol. Animals were sacrificed by overdose of pentobarbital intravenously. Eye tissues were dissected out and kept in 10% formalin for histological examinations.

Results:

No mortality and treatment related clinical sign were noted. Macroscopic examination of the eye showed corneal haze and edema in some animals beginning on day 3 in both 1.5 and 3.0% levofloxacin treated animals. Corneal haze was present on day 11. However, edematous reactions were not present on day 11. Pachymetry data showed an increase in the corneal thickness in the 3% levofloxacin treated right eye compared to the untreated eye. The increase was most evident on days 3, 5, 7, 9 and 10. The sponsor stated that the reading could not be taken on day 2.

The pachymetry data for the right and left eyes of animals in 3% levofloxacin group are shown in the table below.

Day	Right eye	Left eye
1	.429	.431
2		
3	.695	.547
4	.527	.505
5	.641	.458
6	.556	.455
7	.623	.443
8	.459	.439
9	.509	.446
10	.476	.441
11	.525	.414

The sponsor stated that on day 3, there was a statistically significant increase in the size of wound in rabbits treated with 3% levofloxacin solution compared to the untreated eye. The Wound condition improved on day 11. Data suggest that the wound healing was delayed at 3% ophthalmic solution of levofloxacin. However, at 1.5% levofloxacin there was no effect on the wound healing. Based on the nonclinical data, it is suggested that ophthalmological examinations be needed as a follow up at the end of the treatment in patients with corneal ulcer. Ophthalmoscopic examinations showed edematous cornea at 3% solution. Corneal edema was not observed at 1.5% solution.

No change in body weight gain due to the treatment was noted.

No definitive histological changes related to the treatment were reported except that was mentioned above.

Conclusion: Ophthalmic solution of levofloxacin at 3% concentration showed corneal edema and delayed the wound healing in heptanol-induced corneal ulcer model in monkeys. These effects were not observed at 1.5% solution.

Pharmacology summary: Acute treatment with 1.5% levofloxacin ophthalmic drops for two days showed an inhibition of growth of Pseudomonas in the rabbit eye. The antibacterial activity of levofloxacin in vitro showed 99.9% inhibition of bacterial colonies and the Pseudomonas strain was more sensitive than Staphylococcus. The in vivo antibacterial activity of levofloxacin at 0.3 to 3.0% concentrations was reviewed under NDA 21-199. Treatment with 3% levofloxacin, a delay in corneal wound healing and corneal edema were observed in the monkey eye.

Pharmacology conclusions:

Levofloxacin 1.5% ophthalmic solution completely inhibited bacterial growth in the Pseudomonas infected rabbit eye in the presence of experimental corneal ulcer. The wound healing was not impaired at this concentration. However, a delay in healing of corneal ulcer and edema was noted in monkey eyes treated with 3% levofloxacin ophthalmic solution.

III. SAFETY PHARMACOLOGY:

The sponsor did not conduct any safety pharmacology experiment. The review of NDA 21-199 indicates that systemic safety concerns from the ocular treatment with 1.5% levofloxacin are minimal.

III. PHARMACOKINETICS/TOXICOKINETICS:

PK parameters:

Ocular bioavailability:

1. Ocular Pharmacokinetics of 1.5% levofloxacin following a single topical dose in the rabbit, Page 376, vol 2, M4

The ocular distribution of a single 50 µL drop of 1.5% levofloxacin solution was investigated in female NZ albino rabbits. The levofloxacin formulation was with and without ———————————. Three animals were allotted for sacrifice at each time point. Cornea and aqueous humor were collected at 30, 60, 120, 240, 360 and 480 minutes after the drug treatment. Levofloxacin concentrations were determined by the HPLC method. Pharmacokinetic parameters were calculated from the mean tissue concentration-time plot. The data are shown in the table below.

Parameter	Cornea, with ———	Cornea, without ———	Aq. Humor, with ———	Aq. Humor, without ———
Cmax (ng/g)	9,522	11,719	1,102	1,240
Tmax (min)	30	30	60	60
T1/2 (hr)	2.09	1.48	1.26	1.20
AUC (ng.hr/g)	24	22,701	3,255	2,772

Data showed no substantial difference between formulations with or without ———. A measurable concentration of levofloxacin was noted in the eye aqueous humor after a single dose.

2. A repeat dose pharmacokinetic study of 1.5% levofloxacin and single dose pharmacokinetic study of 0.3% ofloxacin in the rabbit eye Page 620 vol 3, M 4.

The study was conducted in NZ albino female rabbits. A drop, 50 µL, levofloxacin or ofloxacin was instilled into both eyes. Animals from groups 1 and 3 were sacrificed at different time

points after the first dose of levofloxacin or ofloxacin. Animals from groups 1 and 2 were sacrificed at several time points up to 12 hours after the last dose. Three animals were allotted for each time point. Cornea and aqueous humor kinetics was determined. The study design is shown in the table below.

Group	Drug	Conc, mg/ml	Percent concentration	Dose
1	Levofloxacin	15	1.5	Q 6h for 48 hr
2	Levofloxacin	15	1.5	Q 4 h for 48 hr
3	Ofloxacin	3	0.3	Once

Levofloxacin and ofloxacin concentrations in the cornea and aqueous humor were determined. The kinetics data after the first and last doses are shown in the table below.

Parameter	Levofloxacin, single dose	Levofloxacin, 48-60 hrs of Q6 hr dosing	Levofloxacin 48-60 hrs of Q4 hr dosing
Cornea			
C_{max} (ng/g)	40781	28839	31657
T_{max} (hr)	0.083	48.083	48.083
T ½ (hr)	1.22	3.12	1.79
AUC $_{0-\infty}$ (ng.hr/g)	35619	38751	36653
Aqueous humor			
C_{max} (ng/ml)	1565	2731	2000
T_{max} (hr)	1.0	49	48.5
$T_{1/2}$ (hr)	0.98	1.58	1.48
AUC $_{0-\infty}$ (ng.hr/ml)	3488	5371	4613

Data showed that levofloxacin did not accumulate in the cornea and aqueous humor after repeated dosing. Kinetic data for Q4 and Q6 hour dosing were not significantly different. The sponsor stated that the lower C_{max} values in the aqueous humor compared to the cornea reflected incomplete transfer of the drug from cornea to the aqueous humor.

3. Pharmacokinetics of topically administered 1.5% levofloxacin in heptanol-wounded vs non-wounded rabbit eyes. Study # PC016-17R, page 1071 vol 4, M4.

The aim of the study was to examine the penetration of the drug from the cornea due to disruption of the corneal epithelium in rabbits. The condition would mimic the corneal ulcerative conditions in humans. The study was conducted following a single ocular instillation of 1.5% levofloxacin ophthalmic solution into the wounded right eye. The right eye was treated with heptanol to induce corneal damage and the left eye served as the control. A total of 30 rabbits were used. Aqueous humor samples were collected at 10, 30, 60, 120 and 240 minutes after the treatment from groups of 6 rabbits per time point. The drug was instilled in 50 µL volume.

Levofloxacin levels in the aqueous humor were determined using liquid chromatographic techniques. The pharmacokinetic parameters are shown in the table below.

Parameter	Intact	Heptanol-wounded
C_{max}, ng/ml	3585	15903
T_{max}, min	120	30
$T_{½}$, hr	1.36	0.83
AUC, ng.hr/ml	11090	20261

Data suggest that penetration of the drug into the anterior chamber of the eye was faster in the wounded cornea compared to the intact cornea. The treatment resulted in the higher exposure to the drug in the eye tissues and it may impact the efficacy and safety of the drug following repeated dosing. Histology data on page 1087, vol 4, module 4 suggest that the heptanol treatment completely denuded the corneal epithelium in most of the rabbits. In addition, a minimal edematous reaction to the connective tissues was noted. Above study was conducted according to the GLP at [redacted]

4. Pharmacokinetics of topically administered 1.5% levofloxacin in mechanically wounded Vs non-wounded rabbit eyes. Page 1306, vol M4, module 4.

The epithelial cells of the cornea were denuded by the laser treatment so that the stromal layer of the cornea was directly exposed to the drug. The bioavailability of the drug in the aqueous humor was compared between the wounded vs non-wounded cornea. The sponsor stated that the experimental conditions represent corneal ulcer in the eye. Animals were anesthetized with ketamine at 35 mg/kg and 5 mg/kg xylazine. Corneal ablation was performed by the laser in the right eye of each rabbit. Two hours after the surgery, a single 50 µL drop of 1.5% levofloxacin was instilled into each eye of the rabbit.

A total of nine rabbits were used in the study. Three rabbits were sacrificed at each time point of 10, 30 and 60 minutes after the drug treatment. Animals were sacrificed by the overdose of sodium pentobarbital, cornea and aqueous humor were collected from both eyes. Histopathological examinations were conducted on the cornea. The aqueous humor levels of the drug were determined by the liquid chromatographic techniques.

The aqueous humor levels of the drug are shown in the table below.

Time	10 min	10 min	30 min	30 min	60 min	60 min
	laser	intact	laser	intact	laser	intact
Mean, ng/ml	5196	2837	6800	5030	3724	5793

The sponsor provided data up to 60 minutes after the treatment. Therefore, exposure levels during longer time were not determined. The data suggest that the drug penetrated the ablated cornea faster than the intact cornea. The sponsor compared the data between heptanol and laser-

induced corneal ablation. Heptanol-induced ablation showed higher levels of levofloxacin in the aqueous humor. The histological data confirmed the removal of cornea in laser treated eyes.

The sponsor stated that both heptanol and laser treatment ablated the cornea in rabbit eyes. However, heptanol represented a better model of experimental corneal ulcer for the levofloxacin study because of improved bioavailability of the drug in the aqueous humor. The study was conducted at [redacted] according to the GLP.

Metabolism: No metabolism study was performed for the NDA

Excretion: No excretion study was performed for the NDA

PK/TK summary:

Several formulations of 1.5% levofloxacin showed bioavailability in the aqueous humor in NZ albino rabbits. Repeated dosing of levofloxacin in the eye did not show increased concentration of the drug in aqueous humor compared to the single dose. Penetration of the drug across the cornea was enhanced when the corneal epithelial cells were denuded by n-heptanol or laser treatment in the rabbit eye. Data suggest that levofloxacin bioavailability in aqueous humor would be greater in corneal ulcer conditions compared to the normal eye.

PK/TK conclusions:

Repeated dosing of 1.5% levofloxacin ophthalmic solution in albino rabbits showed bioavailability of the drug in aqueous humor. The concentration of levofloxacin in aqueous humor was not different between single and repeated doses.

IV. GENERAL TOXICOLOGY:

Study title: 28-day repeat dose toxicity study of levofloxacin in the rabbit eye.

Key study findings: Ophthalmic solution of 3.0% levofloxacin did not show any ocular toxicity in rabbits.

Study no: PC016-07R
Volume # 10, Module 4, and page #: 3059
Conducting laboratory and location: [redacted]

Date of study initiation: March 19, 1999
GLP compliance: Yes
QA report: yes (x) no ( )
Drug, lot # , radiolabel, and % purity: Purity of the drug substance was not provided in the report.
Formulation/vehicle:

Reviewer Name: Asoke Mukherjee, Ph.D. NDA No.: 21-571

Levofloxacin placebo, 1.5% and 3.0%

Ingredient	Placebo, Lot # D99010	1.5%, Lot # D99009	3.0%, Lot # D99011
Levofloxacin		1.5%	3%
Glycerin			
HCl			
NaOH			
Purified water			

The analysis of the ophthalmic solution showed — and — mg/ml of levofloxacin for 1.5 and 3.0% solutions, respectively.

Methods (unique aspects):

Dosing:

Species/strain: male and female Dutch belted rabbits

#/sex/group or time point (main study): Treatment groups are shown in the table below.

Group	Dose	Male	Female
1	Vehicle	5	5
2	1.5%	5	5
3	3.0%	5	5

Satellite groups used for toxicokinetics or recovery: Nil

Age: Approximately 10 months old

Weight: 1.9 –2.6 kg for males and 1.9 –2.8 kg for females

Doses in administered units: A drop of 50 µL of the drug or vehicle solution was instilled into the right eye. One drop every two-hour interval for 6 doses/day. The left eye was untreated control.

Route, form, volume, and infusion rate: Ophthalmic drops

Observations and times:

Clinical signs: Clinical signs and mortality were observed at prestudy and once daily during the treatment period.

Body weights: Body weights were recorded at predose and once a week.

Food consumption: Daily food intake was recorded from one week before the dosing.

Ophthalmoscopy: Eyes were observed for macroscopic changes once a day before the first dose. Slit lamp and indirect ophthalmoscopic examinations were conducted during the pretreatment and once prior to the necropsy. Pupils were dilated with a mydriatic agent before examinations of the posterior chamber of the eye. Eye examinations were also conducted with or without fluorescein. IOP was measured at pretreatment and once a week during the treatment.

Hematology: Blood samples were collected from the auricular vein before the treatment and before necropsy for the determination of hematological and coagulation parameters. Animals were fasted overnight before the blood collection.

Clinical chemistry: Serum chemistry was conducted from the blood collected for the hematology.

Gross pathology: Animals were sacrificed on day 29 by intravenous sodium pentobarbitone overdose. Gross examinations were conducted for the external surface and visceral organs.

Organs weighed: Weights of following organs were recorded:

adrenals, brain, heart, kidneys, liver, ovaries, pituitary, spleen, thymus, thyroid and parathyroid.

Histopathology: Protocol specific tissues were fixed in 10% formalin. Tissues with gross changes and eye tissues were processed for the histological examination.

Toxicokinetics: Blood samples were collected at 30 min after the last dose on days 1, 14 and 28 from 3 animals/sex of groups 2 and 3 animals. Plasma levels of levofloxacin levels were determined by the liquid chromatographic analysis.

Results:

Mortality: No mortality was observed due to the treatment.

Clinical signs: No clinical sign was observed due to the treatment.

Body weights: There was no treatment-related effect on the body weight gain.

Food consumption: There was no treatment-related effect on the food consumption.

Ophthalmoscopy: Macroscopic examination did not show any erythema or edema due to the treatment in the eye. No other ophthalmic changes were noted following slit lamp or ophthalmoscopic examination. The sponsor stated that dystrophy of corneal epithelial was noted in the pretreatment and terminal examination in some animals that was considered to be normal for the rabbit population. Tonometry data did not show treatment-related changes in the IOP.

Hematology:

No treatment related changes were observed in the hematology and coagulation parameters.

Clinical chemistry:

No treatment related changes in the clinical chemistry parameters were noted.

Organ weights:

No treatment related changes were observed in the organ weight.

Gross pathology: No gross pathological changes were noted in the study.

Histopathology:

The histology report (page 3078, vol 10 and M 4) suggested that there was no histological changes related to the treatment.

Toxicokinetics: Plasma concentration (ng/ml) vs day is provided on pages 3182 and 3183, vol 10 and M 4. Data did not show any gender difference in the plasma levels in rabbits. Also, the plasma concentrations were almost similar between days of sampling. The average plasma levels were 67 and 108 ng/ml at 30 min following dosing with 1.5 and 3% levofloxacin, respectively.

Summary of individual study findings: Treatment with 1.5 and 3% ophthalmic solutions for 28 days did not show any ocular toxicity in rabbits. A separate study report for 7-day ophthalmic dosing in rabbits (PC016-05R, page 2917, vol 10 and M 4) also reported a similar finding.

Toxicology summary: Ophthalmic solutions at 1.5% and 3.0% of levofloxacin did not show any ocular toxicity and systemic toxicity in rabbits when treated for 28 days.

Toxicology conclusions: Treatment of 3.0% ophthalmic solution of levofloxacin is safe to rabbit eyes up to 28 days.

IV. GENETIC TOXICOLOGY:

No genetic toxicity report was submitted in the NDA.

VI. CARCINOGENICITY:

No carcinogenicity data were submitted in the NDA.

VII. REPRODUCTIVE AND DEVELOPMENTAL TOXICOLOGY:

No reproductive safety study was submitted in the NDA.

VIII. SPECIAL TOXICOLOGY STUDIES: NIL

IX. DETAILED CONCLUSIONS AND RECOMMENDATIONS:

Conclusions:

Preservative free 1.5% ophthalmic solutions of levofloxacin have been developed for the treatment of infectious corneal ulcer in the eye. Several nonclinical pharmacodynamic studies showed the antimicrobial effect of the drug in experimental corneal ulcer in the rabbits. The data suggest that levofloxacin is more effective for the gram-negative microorganism compared to the gram-positive microorganism. The effect of 1.5% and 3% ophthalmic solutions of levofloxacin on the healing of corneal ulcer was compared. The 3% solution delayed the healing of experimental corneal ulcer in the monkey model. However, 1.5% solution did not show any adverse effect on the healing of corneal ulcer. Repeat dose ocular toxicity studies up to 28 days in Dutch belted rabbits did not show any ocular and systemic toxicity. A previous review for NDA 21-199 of 0.5% levofloxacin solution dated July 3, 2003 also showed that repeated treatment of male Dutch belted rabbits with levofloxacin up to 3.0% was devoid of ocular and systemic toxicity. On the basis of the nonclinical data, the NDA is recommended for approval with minor modifications of labeling as stated below.

General Toxicology Issues: Prolonged treatment with 1.5% levofloxacin could delay the healing of corneal ulcer.

Recommendations: The NDA is approvable on the basis of the nonclinical data.

Labeling with basis for findings: The labeling recommendations are as follows:

Recommendations on Labeling: A proposed label of the product was provided(Page M1-vol 01, 052). The carcinogenicity, mutagenicity, impairment of fertility, and pregnancy information of 1.5% levofloxacin is similar to that already approved for NDA 21-199

(0.5% levofloxacin). However, animal to human dose ratios need to be modified based on the recommended dose of the product.

The ophthalmic medical team leader indicated that the doses of the proposed label should

For carcinogenicity, mutagenicity, impairment of fertility:

In a long term carcinogenicity study in rats, levofloxacin exhibited no carcinogenic potential following daily dietary administration for 2 years; the highest dose (100 mg/kg/day was **100** times the highest recommended human ophthalmic dose.

The mutagenicity portion of the label should be same as proposed.

Levofloxacin caused no impairment of fertility or reproduction in rats at oral doses as high as 360 mg/kg corresponding to **400** times the highest recommended human ophthalmic dose.

Pregnancy:

Teratogenic effects. Pregnancy category C:

Levofloxacin at oral doses of 810 mg/kg/day in rats, which corresponds to approximately **1000** times the highest recommended human ophthalmic dose, caused decreased fetal body weight and increased fetal mortality.

No teratogenic effect was observed when rabbits were dosed orally as high as 50 mg/kg/day, which corresponds to approximately **60** times the highest recommended maximum human ophthalmic dose, or when dosed intravenously as high as 25 mg/kg/day, corresponding to approximately **30** times the highest recommended human ophthalmic dose.

X. APPENDIX/ATTACHMENTS: NIL

Addendum to review: Nil

Other relevant materials (Studies not reviewed, appended consults, etc.):

A written review of following studies has not been provided in this review.

Any compliance issues: Nil

C.C:

Orig NDA # 21-571
HFD-Div File
HFD-550/Pharmacology Reviewer/ A. Mukherjee
HFD-550/Chemist/ S. Khorshidi
HFD-550/Medical Officer/Lucious Lim
HFD-550/CSO/L.Gorski
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