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Efficacy and safety of lomefloxacin on bacterial extraocular disease in the horse

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ABSTRACT. Lomefloxacin is a broad-spectrum fluoroquinolone antibiotic used for the treatment of bacterial extraocular disease. This study investigated the efficacy and safety of lomefloxacin eye drops for bacterial extraocular disease in horses. Lomefloxacin ophthalmic solution (0.3%) was instilled three times daily for 2–5 days in 65 horses diagnosed with bacterial extraocular disease based on clinical findings. Clinical observations and bacteriological examinations were performed at the start of treatment, 2 and 5 days after the start of treatment, and at the discontinuation or termination of treatment. Of the 65 horses, 64 were positive for bacteria, and 22 bacterial genera and 47 bacterial species were identified. The efficacy of lomefloxacin was evaluated in 63 horses; one horse with a negative culture and another with suspected bacterial contamination were excluded. Lomefloxacin was considered to be clinically effective in 54 horses. The major bacterial species identified were *Staphylococcus aureus*, *Streptococcus equi* subsp. *zooepidemicus, Acinetobacter lwoffii*, *Staphylococcus xylosus*, *Staphylococcus vitulinus*, *Enterobacter agglomerans*, *Flavimonas oryzihabitans* and *Staphylococcus sciuri*, with a cumulative disappearance rate of 80% or more at the termination of instillation. Excluding one horse that did not undergo a bacteriological examination, the remaining 62 horses were assessed for bacteriological outcome. Full or partial bacterial clearance was detected in 95% or more of the 62 horses. One of the 65 horses reported adverse events that had no causal relation with the eye drops. Our results showed that lomefloxacin is safe and effective for the treatment of bacterial extraocular disease in horses.

KEY WORDS: bacterial culture, bacterial extraocular disease, clinical study, equine, lomefloxacin

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Bacterial extraocular disease, including conjunctivitis and keratitis, is a common ophthalmic disorder seen in the horse in clinical practice. Local bacterial infection is considered to be one of the causes, and treatment with antibiotics is effective [5, 16, 22]. While it is ideal to select an antibiotic based on the results of bacterial identification and antibiotic sensitivity assessments, this process takes time and many equine veterinarians know that bacterial extraocular disease is primarily caused by *Streptococcus*, *Staphylococcus* or *Pseudomonas* species [13]. Therefore, fluoroquinolone antibiotics (especially ofloxacin), which have a broad antibacterial spectrum, are commonly used as first-choice drugs in Japan [25].

Lomefloxacin is a synthetic antibiotic belonging to the fluoroquinolone group of drugs. Fluoroquinolones inhibit DNA synthesis by inhibiting bacterial DNA gyrase [18] and are effective against gram-positive and gram-negative bacteria, and some anaerobic bacteria [1, 7, 9]. Lomefloxacin is reported to show excellent corneal penetration into the anterior chamber [4, 19], prolonged retention in tears [3, 17] and minimal eye irritation [21], compared to ofloxacin. Lomefloxacin has been reported to be safe and effective for treating bacterial extraocular disease in humans [2, 8], but has not been evaluated in horses. In this study, we investigated the efficacy and safety of lomefloxacin eye drops on bacterial extraocular disease in horses.

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MATERIALS AND METHODS

Sixty-five horses diagnosed with bacterial extraocular disease (bacterial conjunctivitis, keratitis and blepharitis) based on clinical findings were included in the study. The horses were an average of 83 ± 101 months of age and weighed an average of 411 ± 176 kg. Most were thoroughbreds (n=61, 93.8%), but some were mixed breeds or half-bloods (n=4, 6.2%). In sex, the male was 29 (44.6%), and the female was 36 (55.4%). Race horses (n=41, 63.1%, including those bred for racing) were most frequently represented, followed by breeding mares (n=11, 16.9%), riding horses (n=10, 15.4%) and then others (n=3, 4.6%, including pets and stallions). The most common extraocular disease was conjunctivitis (n=38, 58.5%), followed by keratitis (n=16, 24.6%), a complication of conjunctivitis and keratitis (n=7, 10.8%), a complication of conjunctivitis and blepharitis (n=2, 3.1%), and a complication of conjunctivitis, keratitis and blepharitis (n=2, 3.1%).

Topical medications: *Three drops*. Three drops (about 150 μl, and the same applies to the following) of 0.3% lomefloxacin ophthalmic solution (Lomewon®; Senju Pharmaceutical Co., Limited, Osaka, Japan) were instilled into an affected eye of the horses 3 times daily (morning, noon and evening) for a minimum of 2 days and a maximum of 5 days (a maximum of 15 instillations). The horses that received the first instillation at noon or evening and the last instillation

Table 1. Scoring system for clinical conditions

Parameter	Score			
	0	1	2	3
1. Lacrimation	None	Slightly observed in the lid margin	Discoloration or wetting in the hair below the inner canthus	Strong discoloration or wetting in the hair below the inner canthus
2. Photophobia	None	Frequent blinking or slight eye closing	Eyelids are closed one-third or more	Eyelids are almost or completely closed
3. Eye pain	None	Looked slightly painful	Looked painful	Looked very painful
4. Eye discharge	None	Slight discharge with moistening of the lids and hairs just adjacent to lids	Discharge with moistening of the lids and hairs just adjacent to lids	Strong discharge with moistening of the lids and hairs just adjacent to lids
5. Bulbar conjunctival hyperemia	None	Slight vasodilation	Obvious vasodilation	Blood vessels are markedly dilated or have a strong red color
6. Palpebral conjunctival hyperemia	None	Slight redness and vasodilation	Obvious redness and vasodilation	Marked redness and vasodilation
7. Palpebral conjunctival edema	None	Slight edema	Obvious swelling	Marked swelling
8. Corneal opacity	None	Slight opacity and anterior chamber visible	Mild opacity and anterior chamber invisible	Details of iris obscured and anterior chamber invisible
9. Corneal ulcer	None	Crater-like irregularity is smaller than 2 mm	Crater-like irregularity is about 2 mm	Crater-like irregularity is larger than 2 mm

at morning or noon on the 6 days after the start of treatment were also included in the study. A negative control group was not included in this study in consideration of animal welfare, ethical and economic issues. The use of other drugs, such as antibiotics or non-steroidal anti-inflammatory drugs, which may affect the efficacy evaluation, was prohibited for 7 days before the start of treatment and during the course of the study.

Bacteriological examinations and evaluations: Bacteriological examinations were performed at the start of treatment, 2 and 5 days after the start of treatment, and at the discontinuation of treatment or termination of the study. Samples for bacteriology from each lesioned part (conjunctiva and/or cornea) were collected into transport media (BD BBL CultureSwab Plus™; Becton, Dickinson and Co., Tokyo, Japan), and bacteriological examinations were performed at the Research Institute for Animal Science in Biochemistry and Toxicology (Sagamihara, Japan). The swab was applied to a blood agar plate and cultured at 37°C for 24 hr. The bacterial colonies on the agar plates were examined by the naked eye, and the colonies were pure-cultured. The pure-cultured bacteria were identified by a commercially available kit (BD BBL Crystal GP™ and ENF™; Dickinson and Co.) after microscopic observation of the Gram-stained bacteria. If no growth was observed on the agar plate after culturing at 37°C for 24 hr, the plate was cultured at 37°C for another 24 hr to observe bacterial growth.

We used the term "disappearance" to define cases where bacteria detected before the start of the study could no longer be detected after treatment, based on the results of bacteriological examination. The term "microbial substitution" defined those cases where bacteria isolated before treatment disappeared after treatment but different species of bacteria were detected. We defined "partial disappearance" as cases where two or more species of bacteria were identified before treatment and some, but not all, disappeared after treatment (including cases where a new species of bacteria was detected after treatment). Lastly, "unchanged" was defined by isolation of the same species of bacteria before and after treatment.

In vitro susceptibility testing: Susceptibility testing was performed using the microbroth dilution method. The bacteria found at the beginning of the study was grown on Trypticase Soy Broth™ (Becton, Dickinson and Co.) for 18 hr at 35°C, and the turbidity was adjusted to that of a 1.0 McFarland standard. This suspension was used to make a final bacterial inoculum concentration of 10^7 cfu/m*l* using Mueller Hinton Broth™ (Becton, Dickinson and Co.). A prepared range of drug concentrations of lomefloxacin was 0.063–128 μg/m*l*. In inoculations of the bacterial suspension, a Microplanter (Sakuma Factory, Tokyo, Japan) was used, and the cultures were incubated at 35°C for 20 hr under an aerobic atmosphere. The minimum inhibitory concentration (MIC) was defined as the lowest concentration of a test antibiotic that completely inhibited visible bacterial growth.

Clinical observations, efficacy and recurrence evaluations: Clinical observations were performed at the start of treatment, 2 and 5 days after the start of treatment, and at the discontinuation of treatment or termination of the study. The clinical conditions affecting the eyes were scored according to Table 1 referred to the scoring system of Kuriyama *et al.* [11], and the sums of the scores were used to classify the condition as mild (4 to 9), moderate (10 to 18) or severe (19 or higher).

To evaluate efficacy, the rate of improvement was calculated from the total clinical score before and after instillation using the following formula: Improvement rate (%)=(A–B)/A × 100, where A=total clinical score before instillation and B=total clinical score after instillation. An improvement rate of 70% or more was clinically considered "effective,"

Table 2. Bacteria (genus) identified at the start of the study and those minimum inhibitory concentration (MIC) values

Genus	Number of horses	MIC (μg/ml)								MIC_{50}	MIC_{90}
		≤0.063	0.125	0.25	0.5	1	2	4	8		
Acinetobacter (2 species)	11 (16.9%)		3	5	1	1			1	0.25	1
Aerococcus (1 species)	1 (1.5%)									not tested	
Agrobacterium (1 species)	1 (1.5%)									not tested	
Bacillus (3 species)	10 (15.4%)	1		7	2					0.25	0.5
Bergeyella (1 species)	1 (1.5%)	1								—	≤0.063
Corynebacterium (2 species)	3 (4.6%)			1		1	1			—	2
Citrobacter (1 species)	1 (1.5%)		1							—	0.125
Enterobacter (5 species)	11 (16.9%)	7	3		1					≤0.063	0.125
Enterococcus (1 species)	1 (1.5%)								1	—	8
Escherichia (3 species)	3 (4.6%)	2	1							—	0.125
Flavimonas (1 species)	6 (9.2%)	1		2			3			0.25	2
Kocuria (1 species)	1 (1.5%)					1				—	1
Kytococcus (1 species)	1 (1.5%)					1				—	1
Pantoea (1 species)	1 (1.5%)	1								—	≤0.063
Pseudomonas (1 species)	1 (1.5%)					1				—	1
Serratia (1 species)	1 (1.5%)				1					—	0.5
Staphylococcus (11 species)[a)]	46 (70.8%)	1		2	11	15	16			1	2
Streptococcus (6 species)	19 (29.2%)	2				1	1	9	6	4	8
Sphingomonas (1 species)	1 (1.5%)			1						—	0.25
Shigella (1 species)	1 (1.5%)					1				—	1
Stomatococcus (1 species)	1 (1.5%)	1								—	≤0.063
Yersinia (1 species)	1 (1.5%)			1						—	0.25
Negative	1 (1.5%)									—	—

a) Not tested in one strain. MIC_{50}=MIC for 50% of tested pathogenic isolates, MIC_{90}=MIC for 90% of tested pathogenic isolates. Bacteria that the number of strains is less than 5, was calculated only MIC_{90}.

and a rate less than 40% was considered "ineffective," according to "Effectiveness evaluation indicator in clinical trial of antibiotic preparation for animal" that the Ministry of Agriculture, Forestry and Fisheries provided [14].

The efficacy rate was calculated from the number of horses eligible for evaluation. The total clinical score and the scores of each parameter were compared before and after instillation. The Wilcoxon signed-rank test was used for statistical analysis with a significance level of two-sided 5%.

To evaluate recurrence of infection, "effective" cases at the time of the last instillation were observed for an additional 5–7 days. "Recurrence" was defined as an increase in the total clinical score by one or more from the time of the last instillation to the time 5–7 days later.

Safety evaluations: The safety of the eye drops was assessed based on the nature and frequency of adverse events [8, 23]. The eye drops were considered to be safe unless there were serious adverse events related to the eye drops or non-serious adverse events that occurred with a frequency that could not be clinically ignored.

RESULTS

In one horse, clinical and bacteriological examinations that were performed immediately after the 6th installation at 1 day after the start of treatment were counted as data obtained at 2 days after the start of treatment. In this case, even if the data obtained at 1 day after the start of treatment are regarded as data obtained at 2 days after the start of treatment, this may not secure a favorable outcome. Thus, the data obtained at 1 day after the start of treatment were adopted as the data obtained at 2 days after the start of treatment. In 3 horses, recurrence evaluation was performed later than the specified date. These data were still included for the reason stated above.

Topical medications: The median number of eye drop instillations per horse was 15 and varied from 5 to 18. Fourteen horses (21.5%) received fewer than 10 instillations; 32 horses (49.2%) received 10–15 instillations, and 19 horses (29.2%) received more than 15 instillations. The duration of instillation was 2–6 days, with a median of 6 days. The instillation period was less than 3 days in 4 horses (6.2%), 3–5 days in 24 horses (36.9%) and 6 days in 37 horses (56.9%). The number and duration of eye drop instillation were excessive in 19 horses, and instillation was missed in 2 horses (in each case, only one installation was missed, and the instillation rate was 80% or greater). We considered all of the data as it had little effect on the efficacy evaluation.

Bacteriological examinations and evaluations: Of the 65 horses, bacteria were isolated from 64; 22 bacterial genera and 47 bacterial species were identified; only one horse had a negative culture (Table 2). In 61 of the 64 horses, *Acinetobacter*, *Bacillus*, *Corynebacterium*, *Enterobacter*, *Escherichia, Pseudomonas*, *Staphylococcus* or *Streptococcus* was detected. In the remaining three horses, *Agrobacterium*, *Kytococcus* or *Enterococcus* was isolated. *Agrobacte-*

Table 3. Results of bacterial (species) examinations of horses for efficacy evaluation

Bacteria	Number detected at the start (n=63)	Disappearance number 2 days later (n=61)	Cumulative disappearance rate 2 days later	Disappearance number after the last instillation (n=62)	Cumulative disappearance rate after the last instillation
Staphylococcus aureus	12	11	91.7%	1	100.0%
Streptococcus equi subsp. *zooepidemicus*	11	7	63.6%	3	90.9%
Acinetobacter lwoffii	8	8	100.0%		100.0%
Staphylococcus xylosus	8	6	75.0%	1	87.5%
Staphylococcus vitulinus	7	6	85.7%		85.7%
Enterobacter agglomerans	6	5	83.3%		83.3%
Flavimonas oryzihabitans	6	6	100.0%		100.0%
Staphylococcus sciuri	6	6	100.0%		100.0%
Bacillus cereus	5	3	60.0%		60.0%
Staphylococcus equorum	5	3	60.0%		60.0%
Bacillus subtilis	4	4	100.0%		100.0%
Streptococcus constellatus	4	3	75.0%		75.0%
Acinetobacter baumannii	3	2	66.7%		66.7%
Staphylococcus kloosii	3	3	100.0%		100.0%
Corynebacterium aquaticum	2	2	100.0%		100.0%
Enterobacter gergoviae	2	2	100.0%		100.0%
Aerococcus urinae	1	1	100.0%		100.0%
Bacillus brevis	1	1	100.0%		100.0%
Bergeyella zoohelcum	1	1	100.0%		100.0%
Citrobacter freundii	1	1	100.0%		100.0%
Corynebacterium pseudodiphtheriticum	1	1	100.0%		100.0%
Escherichia coli	1	1	100.0%		100.0%
Enterobacter cancerogenus	1	1	100.0%		100.0%
Enterobacter cloacae	1	1	100.0%		100.0%
Enterobacter hafniae	1	1	100.0%		100.0%
Enterococcus gallinarum	1	1	100.0%		100.0%
Escherichia adecarboxylata	1	1	100.0%		100.0%
Escherichia vulneris	1	1	100.0%		100.0%
Kocuria roseus	1	1	100.0%		100.0%
Kytococcus sedentarius	1	1	100.0%		100.0%
Pantoea agglomerans	1	1	100.0%		100.0%
Pseudomonas aeruginosa	1		0.0%	1	100.0%
Serratia marcescens	1	1	100.0%		100.0%
Shigella sp.	1	1	100.0%		100.0%
Sphingomonas paucimobilis	1	1	100.0%		100.0%
Staphylococcus gallinarum	1		0.0%	1	100.0%
Staphylococcus intermedius	1	1	100.0%		100.0%
Staphylococcus lugdunensis	1	1	100.0%		100.0%
Staphylococcus saccharolyticus	1	1	100.0%		100.0%
Staphylococcus simulans	1	1	100.0%		100.0%
Stomatococcus mucilaginosus	1	1	100.0%		100.0%
Streptococcus agalactiae	1	1	100.0%		100.0%
Streptococcus dysgalactiae	1		0.0%	1	100.0%
Streptococcus porcinus	1	1	100.0%		100.0%
Streptococcus uberis	1	1	100.0%		100.0%
Yersinia pseudotuberculosis	1	1	100.0%		100.0%

rium was detected in one horse at the start of treatment, but because *Agrobacterium* is a plant-infecting-microorganism [27], we assumed this was caused by contamination at the time of collection. We excluded this case and the case with the negative culture; a total of 63 horses were included in the study. Both of the 2 excluded cases were of thoroughbred racehorses with conjunctivitis.

The major bacterial species identified in the samples were *Staphylococcus aureus*, *Streptococcus equi* subsp. *zooepidemicus, Acinetobacter lwoffii*, *Staphylococcus xylosus*, *Staphylococcus vitulinus*, *Enterobacter agglomerans*, *Flavimonas oryzihabitans* and *Staphylococcus sciuri*, with a cumulative disappearance rate of 80% or more at the termination of instillation (Table 3).

Table 4. Total clinical score and efficacy rate of the horses for efficacy evaluation

Parameter		At the start	At 2 days after the start	After the last instillation
Clinical score	Range	4–24	0–25	0–23
	Median	14	5	1
P value			≤0.001	≤0.001
Severity[a)]	Severe	10 (15.9%)	3 (4.8%)	2 (3.2%)
	Moderate	34 (54.0%)	10 (15.9%)	1 (1.6%)
	Mild	19 (30.2%)	50 (79.3%)	60 (95.2%)
Improvement rate (%)	Range		–4.2 to 100	4.2 to 100
	Mean (±SD)		58.1 ± 28.9	84.1 ± 19.9
Efficacy rate			38.1% (24/63)	85.7% (54/63)

a) Total clinical score is 4 to 9 for mild, 10 to 18 for moderate and 19 to 24 for severe.

Table 5. Score of clinical conditions by parameter

	Parameter	Lacrimation	Photophobia	Eye pain	Eye discharge	Bulbar conjunctival hyperemia	Palpebral conjunctiva		Cornea	
							Hyperemia	Edema	Opacity	Ulcer
At the start	Range	0–3	0–3	0–3	0–3	0–3	0–3	0–3	0–3	0–3
	Median	3	1	1	2	2	2	1	0	0
At 2 days after the start	Range	0–3	0–3	0–3	0–3	0–3	0–3	0–3	0–3	0–2
	Median	1	0	0	1	1	1	0	0	0
	P value	≤0.001	≤0.001	≤0.001	≤0.001	≤0.001	≤0.001	≤0.001	0.041	0.043
After the last instillation	Range	0–3	0–3	0–3	0–3	0–3	0–3	0–3	0–3	0–2
	Median	0	0	0	0	0	0	0	0	0
	P value	≤0.001	≤0.001	≤0.001	≤0.001	≤0.001	≤0.001	≤0.001	0.009	0.075

Excluding one horse that did not undergo a bacteriological examination after the last instillation, the remaining 62 horses were assessed for the bacteriological outcome. "Disappearance" of bacteria was observed in only four horses (6.5%), and "unchanged" was 3 (4.8%). But, when we also included "microbial substitution (n=35, 56.5%)" and "partial disappearance (n=20, 32.3%)," all or some of the bacteria found at the beginning of the study were not detected in 95% or more of the horses.

In vitro susceptibility testing: Table 2 shows the activity of lomefloxacin against the bacteria found at the beginning of the study. MIC values were 8 μg/m*l* or less, and MIC values for 90% of tested pathogenic isolates (MIC_{90}) were also similar.

Clinical observations, efficacy and recurrence evaluations: Total clinical scores, and improvement and efficacy rates, for efficacy evaluation are shown in Table 4. The total clinical scores at 2 days after the start of treatment and after the last instillation were lower than those at the start of the study, with a significant difference. After the last instillation, the treatment was considered to be effective in 54 horses; no effect was noted in only 2 horses. The scores of all clinical parameters were significantly lower at 2 days after the start of treatment and after the last instillation than at the beginning of the study, except for corneal ulcer (Table 5). The score of corneal ulcer was significantly lower at 2 days after the start of treatment and lower after the last instillation than at the beginning of the study.

Improvement rates were 94.4% (34/36) for conjunctivitis; 75.0% (12/16) for keratitis; 72.7% (8/11) for complications of two or more extraocular diseases; 89.5% (17/19) for mild cases; 88.2% (30/34) for moderate cases; and 70.0% (7/10) for severe cases.

Forty-nine of 54 horses in which the treatment was considered to be effective at the last instillation were evaluated for recurrence. Infection was considered to have recurred in only one horse (2.0%).

Safety evaluations: All 65 horses were assessed for drug safety, and an adverse event (palpebral swelling) was observed in one horse at 1 day after the start of treatment. This horse had a lacerated eyelid wound sutured at the start of the study. Although eye drop instillations were continued after the adverse event, disappearance of swelling was confirmed at the termination of treatment.

DISCUSSION

In horses, extraocular disease, such as conjunctivitis or keratitis, is caused by bacterial infection or migration of foreign bodies [5, 25]. In a bacteriological survey of equine extraocular disease, *Streptococcus*, *Staphylococcus* and *Pseudomonas* were reported to be most commonly detected [13]. In this study, *Pseudomonas* was infrequently detected, but *Staphylococcus* and *Streptococcus* were frequently found. Equine bacterial extraocular disease can also be caused by many other species of bacteria, including *Enterobacter*, *Acinetobacter*, *Bacillus*, *Escherichia* and *Corynebacterium* [13, 15, 24]. In this study, these species as

well as *Staphylococcus* and *Streptococcus* were detected in most cases. Therefore, the cases included in this study were considered to be representative of the general clinical and bacteriological conditions found in a natural environment during the course of the study.

The major species of bacteria detected at the onset of instillation were *Staphylococcus aureus, Streptococcus equi* subsp. *zooepidemicus, Acinetobacter lwoffii, Staphylococcus xylosus, Staphylococcus vitulinus, Enterobacter agglomerans, Flavimonas oryzihabitans* and *Staphylococcus sciuri.* The cumulative disappearance rate at the last instillation was 80% or greater, which suggests that lomefloxacin is effective for these species of bacteria. In a few cases, bacteria disappeared after the last instillation, but all or some of bacteria found at the start of the study were no longer detected in 95% or more of the horses, including cases showing "microbial substitution" and "partial disappearance." These results demonstrate the bacteriological efficacy of lomefloxacin.

The MIC_{90} values of lomefloxacin against the bacteria found at the beginning of the study were 8 μg/m*l* or less (Table 2). Although pharmacokinetic studies in equids are lacking, it has recently been shown that the finding in tear drug concentrations of 1 topical application of 0.3% ciprofloxacin is similar to results in humans and rabbits [6]. If equine pharmacokinetic of lomefloxacin also is similar to that of rabbits, 1 topical application of 0.3% lomefloxacin is considered to result in tear drug concentrations higher than or on the same level with the MIC_{90} for the bacteria found at the beginning of the study for as long as 6 hr after administration [17].

For efficacy evaluation, an ineffective response was found in only 2 horses. One of them had severe keratitis, and *Staphylococcus aureus* was isolated at the start of the study. The veterinarian in charge decided that the treatment was ineffective after performing instillation 6 times in 2 days, and the eye drops were discontinued. In this case, the improvement rate of the total clinical score at 2 days after the start of treatment was as low as 9.1%, but *Staphylococcus aureus* disappeared from the eye. The other horse had a complication of conjunctivitis and keratitis, with severe corneal opacity and a corneal ulcer. *Pseudomonas aeruginosa* was isolated initially. In this case, the eye drops were instilled 14 times in 5 days, but almost no change was observed in the total clinical score (4.2%). However, *Pseudomonas aeruginosa* disappeared after the last instillation. For the treatment of keratitis, adjunct therapy, such as administration of a collagenase inhibitor or non-steroidal anti-inflammatory drug, in addition to antibacterial eye drops is indicated [26], but the use of these drugs was limited for this study. In these cases, the bacteria (the possible cause of the extraocular disease) disappeared after instillation of lomefloxacin; if the adjunct therapy had been used concurrently, the clinical condition score might have improved.

Clinical condition scoring is shown in Table 5. Except for corneal ulcer, the clinical scores of all parameters improved by the last instillation of the eye drops. Treatment of corneal ulcer with instillation of antibiotic eye drops alone did not seem adequate.

The efficacy rate for extraocular disease was 94.4% for conjunctivitis, 75.0% for keratitis and 72.7% for complications of two or more extraocular diseases. Based on these results, the effectiveness of lomefloxacin for these extraocular diseases was confirmed. The efficacy rate for keratitis was rather low compared with that for conjunctivitis, but this is probably because we limited the use of adjunct therapy for keratitis treatment during the study. For complications of 2 or more extraocular diseases, the efficacy rate was also low. This may be explained by the same reason as the above, since keratitis occurred in 9 of 11 horses.

In terms of severity, the efficacy rate was almost 90% for mild and moderate cases and 70% for severe cases, demonstrating that lomefloxacin was effective even in cases of severe extraocular disease. The reason the efficacy rate was lower in severe cases was because keratitis accounted for 70% (7/10) of the severe cases but only 34.0% (18/53) of the mild and moderate cases. Our clinical scores may also have appropriately reflected the clinical conditions.

Of the 54 horses in which the eye drops were considered "effective" at the last instillation, 49 underwent clinical evaluation for recurrence, and recurrence was noted in only 1 horse. The total clinical score of this horse was increased by only 1 between the last instillation and the onset of recurrence, without significant worsening of clinical conditions. These results suggest that the recurrence rate after treatment with lomefloxacin is very low.

Regarding safety, one adverse event was noted in 1 horse, but it was not related to lomefloxacin, suggesting that lomefloxacin eye drops are safe.

In this study, we diagnosed the horses as bacterial extraocular disease with clinical findings and bacteriological examinations, but cytology was not performed. Therefore, it is necessary to evaluate bacteria found at the start of the study as isolation bacteria. However, accompanied by the start of treatment, clinical conditions improved and all or some of bacteria found at the start of the study were no longer detected in most of cases. Moreover, most bacteria found at the start of the study were reported to have the possibility of causing equine extraocular disease [10, 13, 15, 20, 24]. These suggest that extraocular disease in the horses was mainly caused by bacterial infection.

Although ofloxacin has been used for the treatment of equine eye disease as a representative antibiotic in Japan [25], it is reported that ofloxacin causes corneal epithelial disorders and a delay in wound healing with frequent instillation [12]. Therefore, it is recommended that ofloxacin should be used for severe cases or after confirming susceptibility against causative bacteria. As far as we know, lomefloxacin has not been reported to have cytotoxicity to extraocular tissues, and *in vitro* experiments with cultured human conjunctival epithelial tissues have shown that lomefloxacin had lower cytotoxicity than ofloxacin [21]. It has been reported that lomefloxacin has similar efficacy for the treatment of bacterial extraocular disease to ofloxacin in humans [23]. In this study, lomefloxacin was used for the treatment of the horses diagnosed with bacterial extraocular disease based on clinical findings and was confirmed to have excellent

efficacy and safety. Based on these results, lomefloxacin is suitable for the treatment of equine bacterial extraocular disease and is expected to be used as a standard drug in clinical practice.

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