Pharmacokinetics of orbifloxacin and its concentration in body fluids and in endometrial tissues of mares

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Abstract

Pharmacokinetics and distribution of orbifloxacin into body fluids and endometrium was studied in 6 mares after intragastric (IG) administration at a single dose rate of 7.5 mg/kg body weight. Orbifloxacin concentrations were serially measured in serum, synovial fluid, peritoneal fluid, urine, cerebrospinal fluid, and endometrial tissues over 24 hours. Minimum inhibitory concentrations of orbifloxacin were determined for 120 equine pathogens over an 11-month period. The mean peak serum concentration (C_{max}) was 2.41 ± 0.30 μg/mL at 1.5 hours after administration and decreased to 0.17 ± 0.01 μg/mL (C_{min}) at 24 hours. The mean elimination half-life ($t_{1/2}$) was 9.06 ± 1.33 hours and area under the serum concentration vs time curve (AUC) was 20.54 ± 1.70 mg·h/L. Highest mean peritoneal fluid concentration was 2.15 ± 0.49 μg/mL at 2 hours. Highest mean synovial fluid concentration was 1.17 ± 0.28 μg/mL at 4 hours. Highest mean urine concentration was 536.67 ± 244.79 μg/mL at 2 hours. Highest mean endometrial concentration was 0.72 ± 0.23 μg/g at 1.5 hours. Mean CSF concentration was 0.46 ± 0.55 μg/mL at 3 hours. The minimum inhibitory concentration of orbifloxacin required to inhibit 90% of isolates (MIC_{90}) ranged from ≤ 0.12 to > 8.0 μg/mL, with gram-negative organisms being more sensitive than gram-positive organisms. Orbifloxacin was uniformly absorbed in the 6 mares and was well distributed into body fluids and endometrial tissue. At a dosage of 7.5 mg/kg once a day, many gram-negative pathogens, such as *Actinobacillus equuli*, *Escherichia coli*, *Pasteurella* spp., and *Salmonella* spp. would be expected to be susceptible to orbifloxacin.

Résumé

La pharmacocinétique et la distribution de l'orbifloxacine dans les fluides corporels et l'endomètre furent étudiées chez six juments après l'administration intragastrique (IG) d'une dose unique du médicament au taux de 7,5 mg/kg de poids corporel. Les concentrations d'orbifloxacine furent mesurées par dilutions sériées du sérum, du liquide synovial, du liquide péritonéal, de l'urine, du liquide céphalo-rachidien (LCR) et du tissu de l'endomètre sur une période de 24 h. Pendant une période de 11 mois, les concentrations minimales inhibitrices d'orbifloxacine envers 120 bactéries pathogènes pour les chevaux furent déterminées. La moyenne du pic de la concentration sérique (C_{max}) était de 2,41 ± 0,30 μg/mL 1,5 h suite à l'administration et diminua jusqu'à 0,17 ± 0,01 μg/mL (C_{min}) à 24 h. La moyenne de la demie-vie d'élimination ($t_{1/2}$) était de 9,06 ± 1,33 h et la surface sous la courbe de la concentration sérique en fonction du temps était de 20,54 ± 1,70 mg·h/L. La concentration moyenne la plus élevée dans le liquide péritonéal était de 2,15 ± 0,49 μg/mL et fut notée 2 h après l'administration du médicament. La concentration moyenne la plus élevée dans le liquide synovial était de 1,17 ± 0,28 μg/mL et fut obtenue après 4 h. La concentration moyenne la plus élevée dans l'urine était de 536,67 ± 244,79 μg/mL après 2 h. La concentration moyenne la plus élevée dans l'endomètre était de 0,72 ± 0,23 μg/mL à 1,5 h. Les concentration moyenne dans le LCR était de 0,46 ± 0,55 μg/mL à 3 h. La concentration minimale inhibitrice d'orbifloxacine nécessaire pour inhiber 90 % des isolats (CMI_{90}) variaient de ≤ 0,12 à > 8 μg/mL, et les bactéries à Gram négatif étaient plus sensibles que les bactéries à Gram positif. L'orbifloxacine fut absorbé de manière uniforme chez les six juments et fut bien distribué dans les liquides corporels et l'endomètre. À un dosage de 7,5 mg/kg sid, on peut supposer que plusieurs bactéries à Gram négatif pathogènes, telles Actinobacillus equuli, E. coli, Pasteurella *spp. et* Salmonella *spp. seraient sensibles à l'orbifloxacine.*

(Traduit par docteur Serge Messier)

Introduction

Fluoroquinolones are becoming an important group of antimicrobials, particularly for the treatment of infections caused by bacteria resistant to more conventional antibiotics (1–3). Pharmacokinetic studies have revealed good tissue penetration following parenteral or oral administration (3,4). These antibacterial agents are effective against most gram-negative bacteria, including *Enterobacteriaceae* and

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Table I. Mean orbifloxacin concentrations in body fluids and endometrium of 6 mares after IG administration of a single dose of 7.5 mg/kg body weight

	Mean (± SD) orbifloxacin concentrations (μg/mL)					
Hours	Serum	Synovial fluid	Peritoneal fluid	Urine	Endometrium (μg/g)	CSF
0.00[a]	0.00	0.00	0.00	0.00	0.00	0.00
0.08	0.25 ± 0.34	—	—	—	—	—
0.17	0.74 ± 0.28	—	—	—	—	—
0.25	1.24 ± 0.47	—	—	—	—	—
0.50	2.11 ± 0.43	—	—	—	—	—
0.75	2.37 ± 0.25	—	—	—	—	—
1	2.37 ± 0.21	0.41 ± 0.24	1.69 ± 0.29	374.98 ± 303.70	—	—
1.5	2.41 ± 0.30	—	—	—	0.72 ± 0.23	—
2	2.26 ± 0.33	1.05 ± 0.70	2.15 ± 0.49	536.67 ± 244.79	—	—
3	1.80 ± 0.14	—	—	—	0.67 ± 0.12	0.46 ± 0.55
4	1.36 ± 0.27	1.17 ± 0.28	1.88 ± 0.42	363.78 ± 236.03	—	—
8	0.83 ± 0.15	0.89 ± 0.38	1.03 ± 0.38	252.16 ± 28.75	—	—
12	0.42 ± 0.12	—	—	—	—	—
24	0.17 ± 0.01	0.33 ± 0.07	0.17 ± 0.04	69.08 ± 40.27	—	—

— = no sample

[a] dose time

otherwise resistant *Pseudomonas* species (1,4). They have lower, but often therapeutically useful, activity against gram-positive aerobes (1,4). Other principal advantages include their bactericidal activity at low tissue concentrations and good penetration into phagocytic cells. They have a large volume of distribution and low binding to plasma proteins. Tissue concentrations are often higher than concurrent serum concentrations (1,2,4–6).

Gram-negative bacterial infections frequently cause serious clinical disease in horses. Isolates from 233 horses with musculoskeletal infection revealed that gram-negative aerobic bacteria were the most common bacterial group identified in these types of infections (7). Gram-negative bacteria are common isolates in equine pleuropneumonia (8), peritonitis (9), endometritis (10), and cholelithiasis (11). Appropriate antimicrobial therapy for gram-negative infections in horses is limited by the lack of economical, efficacious, safe, and easily administered drugs.

Systemic fluoroquinolone administration at greater than 5 times the manufacturer's recommended dosage produced cartilage lesions in juvenile rats, dogs, guinea pigs, rabbits, and non-human primates (12–17). Articular cartilage damage has also been observed in foals after treatment with enrofloxacin at 10 mg/kg, PO, q24h, for 8 treatments, beginning at 2 wk of age (18). However, the same investigators suggested that a dosage of enrofloxacin of 2.5 to 10 mg/kg, IV, q24h, may be effective in foals, depending on the minimum inhibitory concentration of the pathogen (19). No signs of physical or musculoskeletal changes were seen in adult horses given enrofloxacin at 5 mg/kg, IV, q24h, for 3 wk (20). Other reported side effects of fluoroquinolones include gastrointestinal tract disturbances and stimulatory central nervous signs (21). Orbifloxacin and other fluoroquinolones are not approved for use in horses. In spite of this, extra-label usage of fluoroquinolones is common in equine practice.

Orbifloxacin is a new synthetic fluoroquinolone antimicrobial drug that could potentially become useful in horses. In Japan, orbifloxacin has been highly successful in the treatment of gastrointestinal and respiratory infections in cattle and swine caused by such pathogens as *Pasteurella* spp., *Actinobacillus pleuropneumonia*, and *Escherichia coli* (22). The drug has shown low bacterial resistance (23). The frequent need for orally administered antimicrobial drugs in horses makes it worthy of consideration. To our knowledge, no one has investigated the pharmacokinetics of this oral product in horses.

The purposes of this study reported here were to determine the pharmacokinetic properties of orbifloxacin in adult mares after a single oral dosage of 7.5 mg/kg and to measure its distribution in the body fluids and endometrial tissue. An additional objective was to determine the minimum inhibitory concentration (MIC) of orbifloxacin for pathogenic bacteria isolated from equine patients in our hospital. The pharmacokinetic data and MIC data would then be used to make dosage recommendations for treatment of clinical patients.

Materials and methods

Animals

Six healthy adult mares of various breeds, weighing between 445 and 584 kg, were used in this study. They were given no other medications for a minimum of 4 wk before or during the experiment. A physical examination and complete blood count were performed on each mare prior to the experiment. A physical examination was also conducted after each experiment. Horses were considered healthy on the basis of these findings. Horses were fed water and bermuda grass hay, ad libitum. Horses were kept in individual stalls during the experiment. The project was approved by the University of Florida Institutional Animal Care and Use Committee.

Single intragastric administration

Crushed orbifloxacin tablets (Orbax, 68 mg; Schering-Plough, Kenilworth, New Jersey, USA) in a suspension of 500 mL of water

Table II. Pharmacokinetic parameters (mean ± SD) of orbifloxacin in 6 mares after IG administration at a single dose of 7.5 mg/kg body weight

Pharmacokinetic value	Mean ± SD
AUC (mg·h/L)	19.80 ± 1.70
$t_{1/2}$ (h)	9.06 ± 1.33
MRT (h)	10.60 ± 1.14
Vd_{area} (L/kg)	5.01 ± 0.99
Vd_{ss} (L/kg)	4.04 ± 0.59
Clearance (L/h·kg)	0.38 ± 0.03

AUC — area under the serum concentration vs time curve; $t_{1/2}$ — elimination half-life; MRT — mean residence time; Vd_{area} — the apparent volume of distribution based on area under the curve; Vd_{ss} — the apparent volume of distribution at steady state

Table III. Predicted steady-state maximum and minimum serum concentrations in mares given orbifloxacin IG at a 24-hour dose interval

Orbifloxacin dose (mg/kg)	$C_{max(ss)}$ (μg/mL)	$C_{min(ss)}$ (μg/mL)	C_{max}/MIC[a]	AUC/MIC[a]
5.0	1.74	0.12	14.5	114.7
7.5	2.59	0.18	21.6	171.2

AUC — area under the serum concentration vs time curve; $C_{max(ss)}$ — maximum serum concentration at steady state; $C_{min(ss)}$ — minimum serum concentration at steady state

[a] MIC is 0.12 μg/mL

were administered to non-fasted horses as a single dose (7.5 mg/kg) by nasogastric tube. Blood, synovial fluid, peritoneal fluid, cerebrospinal fluid (CSF), endometrial tissue, and urine specimens were obtained as described (24). Each were collected according to a schedule (Table I). One of the mares did not yield any peritoneal fluid. Endometrial tissue samples were obtained using an endometrial biopsy forcep (Narco Pilling, Fort Washington, Pennsylvania, USA). The CSF sample was obtained from the lumbosacral space by using a 17.7 cm × 18-gauge spinal needle with stylet (Sherwood Medical, St. Louis, Missouri, USA). Of the 6 sample collection attempts, 5 were successful in obtaining CSF with no gross blood contamination.

Preparation of samples

Blood samples were allowed to clot. Serum and body fluid specimens were centrifuged at 2000 × *g* for 10 min, and the supernatant decanted. Serum and body fluid specimens were stored in polypropylene tubes. Supernatant and endometrial biopsy specimens were frozen at −70°C until assayed. Endometrial samples were thawed, weighed, homogenized in a tissue grinder (TenBroeck tissue grinder; Fisher Scientific, Pittsburgh, Pennsylvania, USA) with a 0.9% NaCl solution, and centrifuged at 2000 × *g* for 10 min. The supernatant was assayed.

Orbifloxacin assay

Concentrations of orbifloxacin were determined using an agar well diffusion microbiological assay with a *Bacillus subtilis* strain (Bacto; Difco Laboratories Inc., Detroit, Michigan, USA) as the assay organism (25). Known standards of orbifloxacin were prepared in normal equine serum (5 μg/mL to 0.15 μg/mL) on the date of each experiment and assayed simultaneously with the samples. The lowest limit of detection of the assay was 0.15 μg orbifloxacin/mL and the coefficient of variation for the assay was approximately 2.6%. None of the samples assayed were below the limit of detection.

Minimum inhibitory concentration of orbifloxacin for equine pathogens

Minimum inhibitory concentrations (MIC) of orbifloxacin were determined for all equine bacterial culture specimens submitted to the microbiology laboratory of the University of Florida Veterinary Medical Teaching Hospital from June 16, 1998 to May 18, 1999, using a microtitration strip (JustOne antimicrobial strip; Trek Diagnostic Systems, Westlake, Ohio, USA). Orbifloxacin concentrations tested ranged from 0.12 to 8.0 μg/mL. The assay was validated once monthly by determining MIC values for the known standard organisms *Staphylococcus aureus* (ATCC29213; Trek Diagnostic Systems) and *Escherichia coli* (ATCC25922; Trek Diagnostic Systems). Minimum inhibitory concentrations required to inhibit growth of 50% of isolates (MIC_{50}) and 90% of isolates (MIC_{90}) were both determined, to give some indication of distribution of MIC range for each organism.

Pharmacokinetic analysis

The following equations were fit to the orbifloxacin concentration vs time data for each experiment for each horse; a weighted, nonlinear fit of the mathematical model to the data was done using a computer algorithm that minimized the sum of the squared deviations (26).

$$\text{IG: } C_t = C_1 \cdot e^{-\lambda_1 \cdot t} + C_2 \cdot e^{-\lambda_2 \cdot t} - (C_1 + C_2) \cdot e^{-\lambda_3 \cdot t},$$

where C_t is the serum drug concentration at time t, *e* is the base of Naperian logarithms; C_1, C_2, λ_1, λ_2 and λ_3 are the model values that are fit to the data; and λ_2 is the elimination rate constant, K_{el}. Elimination half-life ($t_{1/2}$) was calculated as the natural logarithm of 2 divided by λ_2. Pharmacokinetic parameters were calculated based on non-compartmental kinetics (27). The area under the serum concentration vs time curve (AUC) was calculated from the model curve using the following equations:

$$AUC_{IG} = C_1/\lambda_1 + C_2/\lambda_2 - (C_1 + C_2)/\lambda_3$$

The area under the moment curve (AUMC) was calculated as:

$$AUMC_{IG} = C_1/\lambda_1^2 + C_2/\lambda_2^2 - (C_1 + C_2)/\lambda_3^2$$

Mean residence time (MRT) was calculated from the following:

$$MRT = AUMC/AUC$$

Volume of distribution based on area under the curve (Vd_{area}) was calculated as follows:

$$Vd_{area}/F = dose/AUC/K_{el}$$

where F is the undetermined bioavailability.

Table IV. Minimum inhibitory concentrations of orbifloxacin and enrofloxacin (μg/mL) required to inhibit growth in 50% (MIC_{50}) and 90% (MIC_{90}) of 120 equine isolates from bacterial submissions at the University of Florida Veterinary Medical Teaching Hospital

		Orbifloxacin			Enrofloxacin		
	n	MIC_{50}	MIC_{90}	Range	MIC_{50}	MIC_{90}	Range
Gram-negative organisms							
Acinetobacter baumannii	1	—	—	0.12	—	—	0.25
Actinobacillus equuli	1	—	—	0.12	—	—	0.25
Aeromonas hydrophilia	1	—	—	0.5	—	—	0.25
Burkholderia cepacia	2	—	—	2.0	—	—	0.5
Citrobacter diversus	2	—	—	0.12	—	—	0.25
Enterobacter cloacae	2	≤ 0.12	≤ 4.0	0.12–4.0	≤ 0.25	≤ 1.0	0.25–1.0
Enterobacter sakazakii	1	—	—	0.12	—	—	0.25
Escherichia coli	22	≤ 0.12	≤ 0.12	0.12–> 8.0	≤ 0.25	≤ 0.25	0.12–2.0
Klebsiella pneumoniae	9	≤ 0.12	≤ 0.25	0.12–0.25	—	—	0.25
Pasteurella pneumotropica	4	—	—	0.12	—	—	0.25
Pasteurella spp.	3	—	—	0.12	—	—	0.25
Proteus vulgaris	1	—	—	0.25	—	—	0.25
Providencia spp.	2	≤ 0.25	≤ 0.5	0.25–0.5	—	—	0.25
Pseudomonas aeruginosa	3	≤ 2.0	≤ 4.0	1.0–4.0	≤ 0.5	≤ 0.5	0.25–0.5
Pseudomonas spp.	2	≤ 1.0	≤ 2.0	1.0–2.0	—	—	0.5
Psychrobacter phenylpyruvica	2	—	—	0.12	—	—	0.25
Salmonella spp.	6	—	—	0.12	—	—	0.25
Gram-positive organisms							
Corynebacterium pseudotuberculosis	1	—	—	0.12	—	—	0.25
Enterococcus spp.	6	≤ 2.0	> 8.0	2.0–> 8.0	≤ 0.5	≤ 2.0	0.5–2.0
Rhodococcus equi	4	≤ 2.0	≤ 4.0	2.0–4.0	≤ 1.0	≤ 2.0	0.5–2.0
Staphylococcus aureus	13	≤ 0.5	≤ 0.5	0.25–1.0	≤ 0.25	≤ 0.25	0.25–1.0
Staphylococcus spp.	1	—	—	0.5	—	—	0.25
Streptococcus equisimilis	5	≤ 2.0	≤ 4.0	2.0–4.0	≤1.0	≤1.0	0.25–1.0
Streptococcus Group D	5	≤ 2.0	≤ 4.0	0.12–4.0	≤1.0	≤1.0	0.25–1.0
Streptococcus zooepidemicus	21	≤ 2.0	≤ 4.0	0.5–4.0	≤1.0	≤1.0	0.5–2.0

—: not applicable; *n*: number of isolates

Volume of distribution at steady state (Vd_{ss}) was determined from:

$$Vd_{ss}/F = dose/AUMC/AUC^2$$

Total serum clearance was calculated from:

$$Clearance/F = dose/AUC$$

Steady-state maximum (C_{max}) and minimum serum orbifloxacin concentrations (C_{min}) were estimated by use of Curry's method, in which maximum and minimum serum concentrations after a single dose are used to predict steady-state values after multiple doses; this method assumes dose-independent kinetics (28).

Results

No adverse affects were observed after orbifloxacin administration in any of the mares. Orbifloxacin was detected in all tissues and body fluid samples from treated mares with the exception of a single synovial fluid and a single urine sample each obtained at 1 h post administration. The mean peak serum concentration was 2.41 ± 0.30 μg/mL at 1.5 h (Table I). Mean serum concentration decreased to 0.17 ± 0.01 μg/mL at 24 h. Mean elimination half-life was 9.06 ± 1.33 h (Table II).

Highest mean synovial orbifloxacin concentration was 1.17 ± 0.28 μg/mL at 4 h. Highest mean peritoneal orbifloxacin concentration was 2.15 ± 0.49 μg/mL at 2 h. Mean concentration of orbifloxacin in peritoneal fluid tended to exceed concurrent synovial fluid concentration except at 24 h after administration. Highest mean urine orbifloxacin concentration was 536.67 ± 244.79 μg/mL at 2 h. Highest mean endometrial orbifloxacin concentration was 0.72 ± 0.23 μg/g at 1.5 h. Mean CSF orbifloxacin concentration at 3 h was 0.46 ± 0.55 μg/mL. At a dosage of 7.5 mg/kg at a 24-hour dose interval, C_{max} and C_{min} at steady state were estimated at 2.59 μg/mL and 0.18 μg/mL, respectively (Table III).

Minimum inhibitory concentrations were determined for a total of 120 isolates. Culture specimens were obtained from sources such as synovial fluid, pleural fluid, transtracheal washes, abscesses, and wounds. The minimum inhibitory concentration of orbifloxacin required to inhibit 90% of isolates (MIC_{90}) ranged from 0.12 to > 8.0 μg/mL, depending on the organism (Table IV).

Discussion

A dosage of 7.5 mg/kg was chosen for the present study based on the oral dosage recommendation of Giguère et al for enrofloxacin (3).

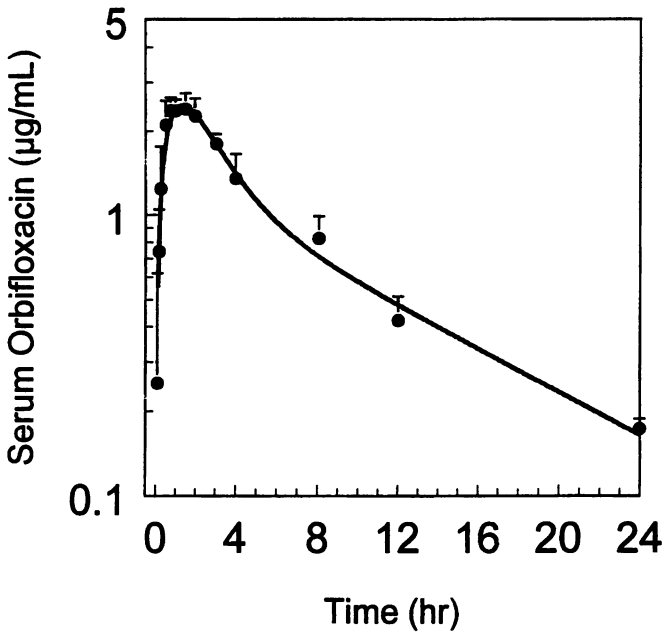

Figure 1. Serum orbifloxacin concentrations (mean ± SD) in mares given a single IG dose of 7.5 mg/kg body weight. The line represents a mathematical model fit to mean concentrations.

Pharmacokinetic values were calculated with the understanding that the amount of absorption of the drug was unknown. An intravenous solution was not available for usage in this study, and as a result, bioavailability could not be determined. Therefore, apparent volume of distribution was expressed as Vd_{ss}/F and clearance was expressed as clearance/F.

This study used a microbiological assay to determine orbifloxacin concentrations. The bioassay does not differentiate between orbifloxacin and its active metabolites. To our knowledge, orbifloxacin and its metabolites have not been investigated in the horse. From a therapeutic point of view, total antimicrobial activity measured by a microbiological assay is adequate to determine a dosage regimen (29). Most investigators evaluating the pharmacodynamic variables correlated to outcome of infection also used a microbiological assay to determine serum concentrations of various fluoroquinolones (30–33).

The relationship between plasma antibiotic concentrations and efficacy is not well defined and often varies between antibiotic families (34). Efficacy of fluoroquinolones against pathogenic bacteria has been evaluated in neutropenic rats and in ill human patients (30,31,35,36). Based on these studies, it has been suggested that $C_{max}/MIC \geq 10$ and $AUC/MIC \geq 125$ are critical breakpoints in determining maximum clinical efficacy (31,35). When these criteria are applied to pathogens with an MIC value of 0.12 µg/mL, $C_{max}/MIC = 20.1$ and $AUC/MIC = 165$. This suggests that our intragastric (IG) dosage of 7.5 mg/kg of orbifloxacin, if given every 24 h, may be adequate for therapy of infections caused by pathogens with an MIC of 0.12 µg/mL. Gram-negative organisms that fit this criteria from table IV included: *Acinetobacter baumannii, Actinobacillus equuli, Citrobacter diversus, Enterobacter sakazakii, Escherichia coli, Pasteurella* spp., *Psychrobacter phenylpyruvica, Salmonella* spp., and the gram-positive isolate *Corynebacterium pseudotuberculosis*. When applied to pathogens with an MIC value of 0.25 µg/mL, $C_{max}/MIC = 9.6$ and $AUC/MIC = 79.2$. This is near the desired ratios of $C_{max}/MIC \geq 10$ and $AUC/MIC \geq 125$ and may be adequate for the successful treatment of pathogens with an MIC value of 0.25 µg/mL. It is possible that an IG dosage of 5.0 mg/kg, q24h, may be adequate for the treatment of infections caused by pathogens with an MIC value of 0.12 µg/mL. At steady state (Table III), $C_{max}/MIC = 14.5$, which exceeds the above criteria for successful outcome. The value for $AUC/MIC = 114.7$, which comes close to a target value of 125. However, to our knowledge, the $C_{max}/MIC \geq 10$ and $AUC/MIC \geq 125$ criteria for maximum therapeutic effect have not been validated in the horse and should be further investigated.

After a single IG administered bolus, mean synovial fluid concentration peaked at 4 h and exceeded serum concentration at 8 and 24 h. Orbifloxacin concentration in synovial fluid exceeded the MIC of most of the gram-negative isolates from our study. However, the MIC_{90} of most gram-positive isolates were ≥ 4.0 µg/mL. These organisms are commonly involved in septic arthritis in adult horses (37). Orbifloxacin may not be efficacious for treating these types of infections.

Urine orbifloxacin concentration was 158 to 406 times higher than concurrent serum concentration, which is consistent with the renal excretion of orbifloxacin or its active metabolite(s), or both. The high urine concentrations suggest that orbifloxacin would be effective for the treatment of several urinary tract pathogens. Mean orbifloxacin peritoneal fluid concentrations peaked at 2 h and exceeded serum concentrations at 4 and 8 h. Mean orbifloxacin peritoneal fluid concentrations were higher than concurrent synovial fluid concentrations except for the sample at 24 h. The majority of horses with a positive peritoneal fluid culture result have polymicrobial infections. *Escherichia coli* was the most common isolate from these samples (9). Orbifloxacin may be efficacious in the treatment of sensitive gram-negative organisms isolated from these infections. The mean CSF concentration was approximately 25% of the concurrent plasma concentration at 3 h. Additional studies are needed to determine orbifloxacin accumulation when the blood-brain barrier is inflamed. Endometrial tissue concentration was 30 and 38% of concurrent plasma concentration at 1.5 and 3 h, respectively. These concentrations of orbifloxacin did not meet the MIC for *Pseudomonas* spp. and *Streptococcus zooepidemicus*. The MIC for *Klebsiella* spp. and *Escherichia coli*, other frequent isolates from mares with endometritis (10), was exceeded. However, the clinical response of endometritis to systemic antibiotics, in general, has not been critically evaluated (10).

Following IG administration, serum concentrations peaked at 2.41 µg/mL of orbifloxacin equivalent activity at 1.5 h. This compares closely to the C_{max} values reported for enrofloxacin of 1.85 (38,39) and 1.88 (40) µg/mL at 45 to 60 min after dosing. However, after IG administration, it appears that serum concentrations of orbifloxacin may be less variable between horses than those observed with enrofloxacin. In an oral dosing study, enrofloxacin tablets (68 mg tablets) were ground into a powder and administered without fasting at a 5 mg/kg single dose (38). Enrofloxacin appeared to be rapidly yet variably absorbed following oral administration to the non-fasted horse (38). When evaluating its pharmacokinetic properties, AUC was the most variable parameter studied with a

coefficient of variation of 76% (38). Another single dosing study, using an enrofloxacin oral solution (32.3 mg/mL) administered without fasting at 7.5 mg/kg reported a coefficient of variation of 29% (39). The coefficient of variation for orbifloxacin in this study was 9%. Perhaps orbifloxacin is more uniformly absorbed in the non-fasted adult horse than enrofloxacin. This may avoid extreme variations in serum concentrations that have been reported with enrofloxacin (38), making orbifloxacin safer than enrofloxacin with respect to potential toxic side effects when administered to horses. This warrants further investigation.

When comparing MIC_{90} concentrations of orbifloxacin with enrofloxacin concentrations required to inhibit the growth of 120 equine isolates (Table IV), orbifloxacin had lower MIC values for specific gram-negative organisms. Some of the more common gram-negative isolates with lower orbifloxacin MIC_{90} concentrations were *Actinobacillus equuli, Escherichia coli, Pasteurella* spp., and *Salmonella* spp. The orbifloxacin MIC_{90} concentration for these specific organisms was 0.12 μg/mL compared to an MIC_{90} of 0.25 μg/mL for enrofloxacin. Orbifloxacin did not appear to demonstrate therapeutically useful activity against gram-positive aerobes, with the exception of *Corynebacterium pseudotuberculosis*, especially when compared to enrofloxacin.

At a dosage of 7.5 mg/kg once a day, many gram-negative infections would be expected to be susceptible to orbifloxacin. However, based on steady state C_{max} and C_{min} calculated by Curry's method, orbifloxacin may be effective for the treatment of susceptible gram-negative infections in horses when administered orally at 5 mg/kg once a day. Although this reduction in dosage would reduce the cost of treatment, multiple dose studies may need to be performed to confirm the effectiveness of this lower dosage. Additional studies may also be necessary to confirm the efficacy and safety of this drug in a clinical setting as well as to evaluate the penetration of orbifloxacin into diseased tissues. Orbifloxacin was uniformly absorbed in the 6 mares in this study. Orbifloxacin was well distributed into body fluids and endometrial tissue. Potential adverse side effects, such as the suspected fluoroquinolone-associated arthropathy in foals, would require further investigation. Until proven otherwise, this drug should be avoided in young growing horses because of the potential for articular cartilage damage.

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