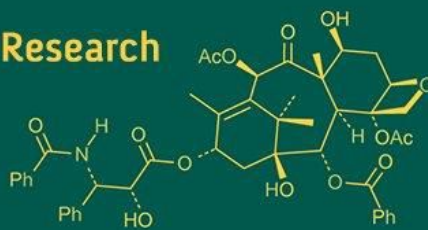


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Pharmacokinetics of danofloxacin following intravenous and intramuscular administration in Kankrej cow calves

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Abstract

The present study evaluated the pharmacokinetics of danofloxacin following single intravenous (IV) and intramuscular (IM) administration at a dose of 5.0 mg/kg body weight in Kankrej cattle calves. After IV administration, plasma concentrations of danofloxacin remained above the minimum inhibitory concentration ($MIC \leq 0.125 \mu\text{g/mL}$) for up to 24 h. The elimination half-life was 5.94 ± 0.83 h, clearance was 0.37 ± 0.01 L/h/kg, area under the plasma concentration-time curve ($AUC_{0-\infty}$) was $13.73 \pm 0.57 \mu\text{g} \cdot \text{h/mL}$, and mean residence time (MRT) was 7.44 ± 1.03 h. Following IM administration, danofloxacin was rapidly absorbed, attaining a peak plasma concentration (C_{max}) of $1.37 \pm 0.08 \mu\text{g/mL}$ at 0.71 ± 0.14 h, and therapeutic concentrations were maintained for approximately 24 h. The elimination half-life (7.22 ± 0.57 h) and MRT (9.90 ± 0.47 h) were slightly prolonged after IM administration, while absolute bioavailability was high (96.83%). Overall, danofloxacin exhibited favorable pharmacokinetic characteristics, including rapid absorption, extensive systemic availability, and sustained plasma exposure. The study provides breed-specific pharmacokinetic information in Kankrej cattle calves and supports rational dose optimization and effective clinical use of danofloxacin in cattle under Indian field conditions. These data contribute valuable reference information for future pharmacodynamic integration and therapeutic recommendations development.

Keywords: Bioavailability, danofloxacin, intramuscular, intravenous, Kankrej cattle calves, pharmacokinetics

Introduction

Danofloxacin is a third-generation fluoroquinolone antibacterial agent developed exclusively for veterinary use and formulated as the mesylate salt (Chen *et al.*, 2023; Zhang *et al.*, 2023) [4, 21]. It exhibits broad-spectrum activity against Gram-negative and Gram-positive bacteria, as well as *Mycoplasma* and *Chlamydia* species (Shem-Tov *et al.*, 1998) [17], and has regulatory approval for veterinary use in food-producing and dairy animals in several countries (EMA, 2002) [6]. Favorable pharmacological properties, including good cellular penetration, wide tissue distribution with preferential accumulation in pulmonary tissues, and sustained concentration-dependent bactericidal activity even at declining plasma concentrations, have made danofloxacin an important molecule of interest for the treatment of respiratory, gastrointestinal, and systemic bacterial diseases in livestock (Aliabadi & Lees, 2001; Giles *et al.*, 1991) [1, 9].

However, to the best of our knowledge, pharmacokinetic data on danofloxacin in Indian cattle breeds are not available. Pharmacokinetic behavior of fluoroquinolones may vary among breeds due to differences in physiological characteristics, metabolic capacity, and management practices, which can influence drug disposition (Toutain *et al.*, 2010) [19]. Indigenous cattle breeds play a central role in India's dairy sector and frequently require antimicrobial therapy. In the absence of breed-specific pharmacokinetic data, rational dose optimization remains challenging. Therefore, the present study aimed to characterize the pharmacokinetics of danofloxacin following single-dose intravenous and intramuscular administration in Kankrej cattle calves to generate breed-specific data for rational dosing and effective clinical use.

Materials and Methods

Experimental location and ethical approval

The animals used in this study were sourced from and housed at the Large Animal Experimental Facility of the Livestock Research Station (LRS), Kamdhenu University, Sardarkrushinagar, Gujarat, India (24.32°N, 72.30°E; 154.5 m above mean sea level). The experimental site lies in the tropical semi-arid climatic zone of North Gujarat. The study was conducted during June and July 2025, with ambient temperatures ranging from 25 °C to 35 °C. The experimental protocol was approved by the Institutional Animal Ethics Committee (IAEC) of the College of Veterinary Science and Animal Husbandry, Kamdhenu University, with protocol Approval No. VETCOLL/IAEC/2024/23/PROTOCOL-01, and all procedures were conducted in accordance with CCSEA guidelines.

Experimental animals

Six healthy male Kankrej cattle calves (*Bos indicus*), aged between 6 and 10 months and weighing 111-165 kg, were used in the pharmacokinetic study. None of the animals had a history of exposure to antimicrobial agents since birth. The animals were housed under standard management conditions and were provided with adequate green fodder, dry roughage, and the required quantity of concentrate feed to meet their daily nutritional requirements. Clean drinking water was available *ad libitum* throughout the experimental period. Prior to initiation of the study, all animals were acclimatized and kept under continuous observation for a period of 10 days to confirm their health status. All necessary management practices were followed to minimize stress and prevent the occurrence of disease.

Chemicals and reagents

Danofloxacin (purity $\geq 98\%$; CAS No. 119478-55-6) was procured from JK Chemicals, Vapi, Gujarat, India. HPLC-grade water, methanol, acetonitrile, formic acid, perchloric acid (70%), and triethylamine were procured from S. D. Fine Chemicals Ltd., Mumbai, India.

Experimental design

The study was conducted in a randomized two-period crossover design in six healthy cattle calves. In Phase I, three calves received danofloxacin intravenously and three intramuscularly. After a 14-day washout period, the routes of administration were crossed over in Phase II, such that each animal received the drug by both routes. This design allowed within-animal comparison of pharmacokinetic parameters.

Drug Administration and Blood Sampling

The calves received a single dose of danofloxacin at a dose rate of 5.0 mg/kg body weight. Intravenous administration was performed via the jugular vein, while intramuscular injections were administered into the neck (trapezius) muscle. An 18-G needle was used for drug administration by both routes. Blood samples were collected into heparinized tubes from a cannulated contralateral jugular vein at 0 min (pre-dosing), 2 min (0.033 h), 5 min (0.083 h), 10 min (0.167 h), 15 min (0.25 h), 30 min (0.5 h), 1, 2, 4, 8, 12, 24, 36, and 48 h following intravenous administration. For intramuscular administration, the 2-minute (0.033 h) sampling time point was excluded, while all other sampling

time points remained the same as those used for the intravenous route. Approximately 2 mL of blood was collected at each sampling time point into heparinized tubes. Plasma was separated by centrifugation at $2856 \times g$ for 5 min. The clear plasma supernatant was carefully transferred into 2 mL microcentrifuge tubes and stored at -20 °C until quantitative analysis of danofloxacin concentration using the UHPLC method.

Extraction of danofloxacin

Danofloxacin was extracted from plasma using a modified method described by Patel *et al.* (2024) [14]. Briefly, a 100 μ L aliquot of plasma was transferred into a 2 mL microcentrifuge tube, and an equal volume (100 μ L) of 20% perchloric acid prepared in ice-cold acetonitrile was added. The samples were vortex-mixed for 2 min and subsequently centrifuged at $11,424 \times g$ for 10 min at 4 °C in a refrigerated centrifuge. The resulting clear supernatant was carefully transferred into a separate microcentrifuge tube, and a 20 μ L aliquot was injected into the UHPLC system for analysis.

UHPLC apparatus

Plasma danofloxacin concentrations were determined using an Ultra High Performance Liquid Chromatography (UHPLC) system (Dionex Ultimate 3000®, Thermo Fisher Scientific, Germany). The system was equipped with a UV detector, a gradient solvent delivery pump, and a manual injector. Chromatographic separation of danofloxacin was carried out using a reverse-phase C18 column (ODS-3V, GL Sciences Inc., Japan; 250×4.6 mm), maintained at room temperature (20-30 °C). A 20 μ L injection loop was used for sample injection, and chromatographic data were acquired and processed using Chromeleon™ software (version 6.8). The total run time for each injection was 15 minutes, and the mean retention time of danofloxacin under these conditions was 5.5 ± 0.4 minutes. The mobile phase consisted of a mixture of 0.01 M formic acid buffer and acetonitrile (79:21, v/v), delivered at a flow rate of 1.0 mL/min. The 0.01 M formic acid buffer was prepared by dissolving 460 μ L of formic acid in 1 L of HPLC-grade water, and the pH was adjusted to 2.8 using 230 μ L of triethylamine. Isocratic elution was employed throughout the analysis, and detection was carried out at a wavelength of 282 nm.

Method validation

The UHPLC method used for the estimation of danofloxacin was validated in-house for linearity, recovery, accuracy, specificity, precision (intra-day and inter-day), limit of detection (LOD), and limit of quantification (LOQ). Method validation was performed in accordance with the guidelines described by the CDER (1994) and ICH (2005). Quantification of danofloxacin in cattle plasma demonstrated linearity over the concentration range of 0.1-12.0 μ g/mL, with a regression coefficient (r^2) of 0.9997. The intra-day and inter-day precision for the three quality control samples was within acceptable limits, with relative standard deviation (RSD) values not exceeding 4.41%. The mean recovery of danofloxacin from cattle plasma was found to be 91%.

Pharmacokinetic analysis

Pharmacokinetic parameters of danofloxacin were estimated using PK Solver 2.0 (Zhang *et al.*, 2010) [22], a Microsoft Excel add-in based on statistical moment theory, employing

a model-independent, non-compartmental approach. Plasma concentration-time data from individual calves following intravenous and intramuscular administration were analyzed without assuming any underlying compartmental model (Gibaldi & Perrier, 1982) [8]. The analysis was based on the area under the plasma concentration-time curve (AUC) and the area under the first moment curve (AUMC), calculated using the linear trapezoidal rule. Pharmacokinetic parameters, including elimination half-life, mean residence time, clearance, and volume of distribution, were derived from the terminal phase characterized by the slope (λ_z). This approach provides a reliable and widely accepted description of drug disposition derived directly from observed concentration-time data. For IM administration, the bioavailability (F) was determined using the following formula

$$F = (AUC_{IM}/AUC_{IV}) \times 100$$

Results and Discussion

The extraction and UHPLC quantification procedures used for danofloxacin demonstrated high specificity and selectivity, with satisfactory accuracy and precision for plasma analysis. Danofloxacin was consistently eluted at a retention time of approximately 5.5 ± 0.4 min, with no interference from endogenous plasma components or impurity peaks (Figure 1). The analytical method exhibited excellent linearity across the concentration range of 0.1–12 $\mu\text{g/mL}$, with a correlation coefficient (R^2) of 0.9997. The recovery of danofloxacin was approximately 91%, as determined in triplicate at concentration levels of 0.1, 1, and 8 $\mu\text{g/mL}$. The intra-day coefficient of variation ranged from 2.33% to 4.41%, while the inter-day coefficient of variation ranged from 1.59% to 3.18%.

All the cattle calves were free from any adverse reaction and exhibited normal feed and water intake following intravenous and intramuscular administration of danofloxacin (5.0 mg/kg body weight).

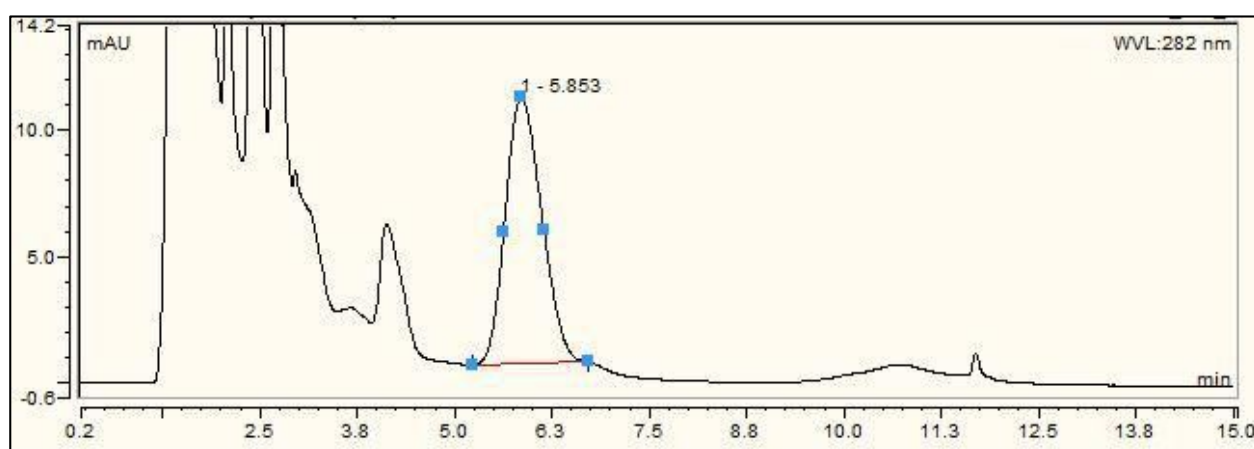


Fig 1: Chromatogram showing the peak of danofloxacin with a retention time of 5.853 minutes.

The plasma danofloxacin concentrations at each sampling time point following intravenous and intramuscular administration at a dose rate of 5.0 mg/kg body weight are presented as mean and standard error (SE) values for six calves in Table 1, and the corresponding superimposed semilogarithmic plot of mean plasma concentration *versus* time is shown in Figure 2.

Following intravenous administration of danofloxacin at 5.0 mg/kg body weight, a rapid decline in plasma drug concentrations was observed during the initial sampling period, indicating a prompt distribution of the drug from the central compartment to peripheral tissues. This early decrease was followed by a more gradual and sustained decline in plasma concentrations, suggesting elimination was predominantly governed by systemic clearance processes. Danofloxacin remained detectable in plasma up to 24 h post-administration, with concentrations exceeding the reported minimum inhibitory concentration ($MIC \leq 0.125 \mu\text{g/mL}$) for most of the dosing interval. The absence of detectable drug levels at 36 and 48 h post-administration reflects effective elimination of danofloxacin from systemic circulation. The observed biphasic decline pattern is consistent with first-order elimination kinetics reported for fluoroquinolones in cattle and indicates rapid tissue distribution followed by elimination via renal and hepatic pathways. Overall, the decline in plasma concentrations demonstrates a favorable pharmacokinetic profile characterized by adequate systemic exposure and sustained

therapeutic concentrations over a 24 h period following intravenous administration.

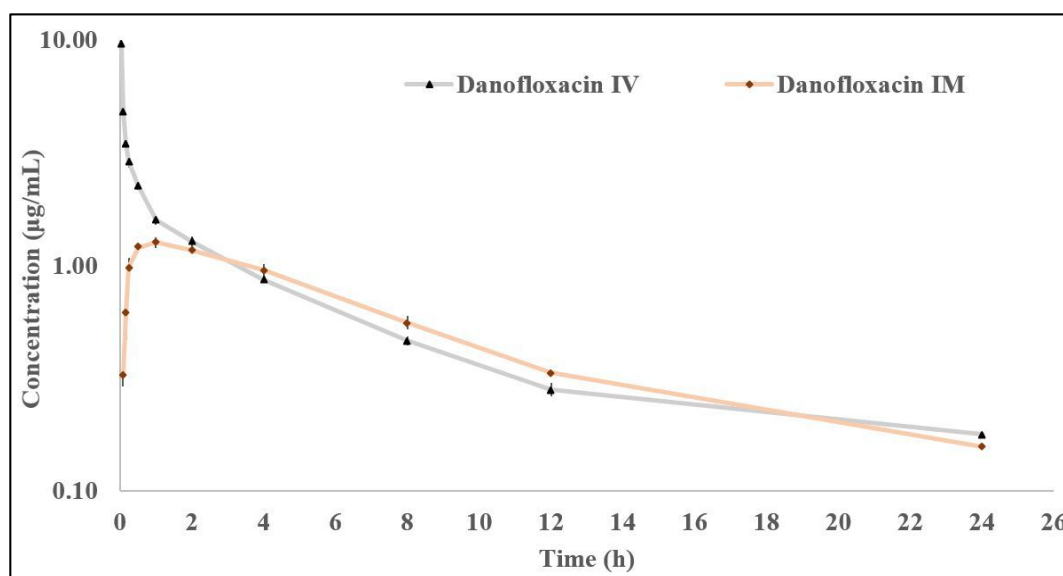
After IV administration of danofloxacin at 5.0 mg/kg body weight in cattle calves, the mean plasma concentration at the first sampling point, 0.033 h (2 minutes), was $9.57 \pm 0.35 \mu\text{g/mL}$. Thereafter, mean plasma concentrations declined progressively and were recorded as 4.81 ± 0.31 , 3.46 ± 0.16 , 2.90 ± 0.18 , 2.26 ± 0.10 , 1.59 ± 0.07 , 1.28 ± 0.07 , 0.86 ± 0.02 , 0.46 ± 0.02 , 0.28 ± 0.02 , and $0.18 \pm 0.00 \mu\text{g/mL}$ at 0.083, 0.167, 0.25, 0.50, 1, 2, 4, 8, 12, and 24 h, respectively. Danofloxacin concentrations were below the limit of detection in all plasma samples collected at 36 and 48 h post-administration.

After IM administration of danofloxacin at the same dose rate (5.0 mg/kg body weight), the mean plasma concentration at the first sampling point, 0.083 h (5 minutes), was $0.33 \pm 0.03 \mu\text{g/mL}$. Plasma concentrations subsequently increased and reached a mean peak plasma concentration (C_{max}) of $1.37 \pm 0.08 \mu\text{g/mL}$ at a mean time to peak concentration (T_{max}) of 0.71 ± 0.14 h. Mean plasma concentrations at subsequent time points of 0.167, 0.25, 0.50, 1, 2, 4, 8, 12, and 24 h were 0.62 ± 0.04 , 0.98 ± 0.14 , 1.20 ± 0.10 , 1.27 ± 0.04 , 1.17 ± 0.07 , 0.95 ± 0.03 , 0.56 ± 0.06 , 0.33 ± 0.04 , and $0.16 \pm 0.01 \mu\text{g/mL}$, respectively. Similar to the intravenous route, plasma concentrations were below the limit of detection in all samples collected at 36 and 48 h following intramuscular administration.

Table 1: Plasma concentrations (Mean $\pm$ SE) of danofloxacin following single dose intravenous and intramuscular administration at the dose rate of 5 mg/kg body weight in cattle calves (n = 6)

Time point (h)	Plasma danofloxacin concentration ($\mu\text{g/mL}$) Mean $\pm$ SE (n = 6)	
	Intravenous	Intramuscular
0.033	9.57 $\pm$ 0.35	-
0.083	4.81 $\pm$ 0.31	0.33 $\pm$ 0.03
0.167	3.46 $\pm$ 0.16	0.62 $\pm$ 0.04
0.25	2.90 $\pm$ 0.18	0.98 $\pm$ 0.14
0.5	2.26 $\pm$ 0.10	1.20 $\pm$ 0.10
1	1.59 $\pm$ 0.07	1.27 $\pm$ 0.04
2	1.28 $\pm$ 0.07	1.17 $\pm$ 0.07
4	0.86 $\pm$ 0.02	0.95 $\pm$ 0.03
8	0.46 $\pm$ 0.02	0.56 $\pm$ 0.06
12	0.28 $\pm$ 0.02	0.33 $\pm$ 0.04
24	0.18 $\pm$ 0.00	0.16 $\pm$ 0.01
36	BDL	
48	BDL	

BDL-Below detection limit

**Fig 2:** Semilogarithmic plot of mean ($\pm$ SE) plasma danofloxacin concentration-time profiles following intravenous and intramuscular administration at a dose of 5.0 mg/kg body weight in cattle calves (n = 6).

In the present study, mean plasma concentrations of danofloxacin following intravenous and intramuscular administration at a dose rate of 5.0 mg/kg body weight in cattle calves were maintained at or above the MIC₉₀ value of

0.125 $\mu\text{g/mL}$ for the majority of veterinary pathogens causing diseases in cattle, as reported by Giles *et al.* (1991)^[9] for approximately 24 h.

Table 2: Pharmacokinetics (PK) parameters of danofloxacin following single dose IV and IM administration (5 mg/kg body weight) in cattle calves (n = 6)

PK Parameters	Unit	Intravenous		Intramuscular	
		Mean $\pm$ SE (n = 6)	Range	Mean $\pm$ SE (n = 6)	Range
β	h^{-1}	0.13 $\pm$ 0.02	0.08-0.17	0.10 $\pm$ 0.01	0.07-0.12
$t_{1/2\beta}$	h	5.94 $\pm$ 0.83	4.04-8.58	7.22 $\pm$ 0.57	5.73-9.60
Cp^0	$\mu\text{g/mL}$	15.20 $\pm$ 0.85	12.52-18.00	-	-
C_{max}	$\mu\text{g/mL}$	-	-	1.37 $\pm$ 0.08	1.17-1.63
T_{max}	h	-	-	0.71 $\pm$ 0.14	0.25-1.00
$\text{AUC}_{0-\infty}$	$\mu\text{g}\cdot\text{h/mL}$	13.73 $\pm$ 0.57	12.58-15.78	13.15 $\pm$ 0.58	11.89-15.64
MRT	h	7.44 $\pm$ 1.03	5.24-10.74	9.90 $\pm$ 0.47	8.40-11.63
$\text{V}_{\text{d(are)}}$	L/kg	3.06 $\pm$ 0.30	2.31-3.97	3.99 $\pm$ 0.36	3.32-5.70
$\text{V}_{\text{d(ss)}}$	L/kg	2.66 $\pm$ 0.25	2.08-3.46	-	-
Cl_B	L/h/kg	0.37 $\pm$ 0.01	0.32-0.40	0.38 $\pm$ 0.02	0.32-0.42
F	%	-	-	96.83	77.04-110.64

(Notations used:- β : Elimination rate constant; $t_{1/2\beta}$: Elimination half-life; Cp^0 : Theoretical concentration of drug in plasma at zero-time; C_{max} : Observed peak plasma concentration; T_{max} : Time at which C_{max} was observed; $\text{AUC}_{0-\infty}$: Area under curve from time zero to infinity; MRT: Mean Resident Time; $\text{V}_{\text{d(ss)}}$: Volume of distribution at steady state; $\text{V}_{\text{d(are)}}$: Apparent volume of distribution; Cl_B : Total body clearance; F: Absolute Bioavailability)

The pharmacokinetic (PK) parameters of danofloxacin following single intravenous and intramuscular administration at a dose rate of 5.0 mg/kg body weight in cattle calves were determined from plasma concentration-time data and are presented in Table 2. Pharmacokinetic analysis was performed using a model-independent, non-compartmental approach, and the results are expressed as mean $\pm$ standard error (SE) for six calves. To the best of our knowledge, pharmacokinetic studies evaluating danofloxacin at a dose rate of 5.0 mg/kg body weight following intravenous and intramuscular administration in either Indian or exotic cattle calves have not been previously reported. In contrast, available pharmacokinetic data in cattle are limited to studies conducted at lower dose levels, such as 1.25 mg/kg body weight in exotic cattle breeds (Aliabadi & Lees, 2003; Giles *et al.*, 1991; Shem-Tov *et al.*, 1998) [2, 9, 17], or at higher dose regimens of approximately 8.0 mg/kg body weight in premature calves (Corum *et al.*, 2019) [5]. Nevertheless, danofloxacin has been pharmacokinetically characterized in a wide range of animal species, providing a useful basis for cross-species comparison and interpretation of the present findings.

In the present study, the mean elimination rate constant (β) values were $0.13 \pm 0.02 \text{ h}^{-1}$ and $0.10 \pm 0.01 \text{ h}^{-1}$ following intravenous and intramuscular administration of danofloxacin, respectively. These values are lower than those previously reported in cattle calves, where β values of 0.17 h^{-1} (IV) and 0.18 h^{-1} (IM) were observed by Giles *et al.* (1991) [9]. In the present study, the elimination half-life ($t_{1/2\beta}$) of danofloxacin was estimated to be $5.94 \pm 0.83 \text{ h}$ and $7.22 \pm 0.57 \text{ h}$ following intravenous and intramuscular administration, respectively. These values are higher than those reported by Aliabadi and Lees (2003) [2], who observed elimination half-lives of 2.65 h (IV) and 4.03 h (IM) in cattle calves. In contrast, markedly longer elimination half-lives of 17.47 h (IV) and 28.41 h (IM) have been reported by Corum *et al.* (2019) [5] in premature calves. These wide variations in the elimination half-life across different animals might be due to the animal's drug metabolizing capacity, protein content, urine and body fluid pH, physiological differences (age of animals, gender), and many other factors (Toutain *et al.*, 2010) [19]. Because elimination half-life ($t_{1/2\beta}$) depends on both clearance (Cl) and volume of distribution (V_d) (Gibaldi & Perrier, 1982) [8], differences in $t_{1/2\beta}$ between studies likely reflect variability in Cl and V_d . The clearance (Cl) values observed in the present study were 0.37 ± 0.01 and $0.38 \pm 0.02 \text{ L/h/kg}$ following intravenous and intramuscular administration, respectively, indicating comparable systemic elimination across both routes. These values are higher than the clearance of 0.18 L/h/kg reported by Corum *et al.* (2019) [5] following intravenous administration in calves, but substantially lower than the markedly elevated clearance of 1.02 L/h/kg documented by Aliabadi and Lees (2003) [2]. In contrast, the clearance obtained in the present study closely aligns with the value of 0.32 L/h/kg reported by Manzoor *et al.* (2017) [12] following intravenous administration in Kundhi buffaloes. Differences in renal capacity among different animals can contribute to variability in drug elimination efficiency. Variations in renal blood flow, glomerular filtration rate, tubular secretion, and tubular reabsorption directly influence the rate at which the drug is cleared from plasma (Toutain & Bousquet-Mélou, 2004) [18]. The steady-state volume of distribution ($V_{d(ss)}$) observed in

the present study is in close agreement with previously reported values of 2.04 L/kg (Aliabadi & Lees, 2003) [2] and 2.48 L/kg (Giles *et al.*, 1991) [9]. Comparable $V_{d(ss)}$ values have also been reported in other animals, including goats (2.66 L/kg ; Sartini *et al.*, 2021) [16] and sheep (2.19 L/kg ; Escudero *et al.*, 2007) [7]. The apparent volume of distribution ($V_{d(area)}$) values in the present study were found to be $3.06 \pm 0.30 \text{ L/kg}$ and $3.99 \pm 0.36 \text{ L/kg}$ following IV and IM administration, respectively. Comparable ($V_{d(area)}$) value of 4.35 L/kg was reported by Sappal *et al.* (2009) [15] after intravenous danofloxacin administration in buffalo calves. The relatively high volume of distribution observed in the present study indicates extensive distribution of danofloxacin into extravascular tissues, which is consistent with the known pharmacokinetic behavior of fluoroquinolones (Zhanel *et al.*, 1999) [20]. The area under the plasma concentration-time curve from time zero to infinity ($AUC_{0-\infty}$) was $13.73 \pm 0.57 \mu\text{g}\cdot\text{h/mL}$ and $13.15 \pm 0.58 \mu\text{g}\cdot\text{h/mL}$ following intravenous and intramuscular administration, respectively. As AUC is directly dependent on the administered dose, substantially lower AUC values of $1.24 \mu\text{g}\cdot\text{h/mL}$ (IV) and $1.42 \mu\text{g}\cdot\text{h/mL}$ (IM) were reported by Aliabadi and Lees (2003) [2] following danofloxacin administration at a lower dose of 1.25 mg/kg in cattle calves. Similarly, Giles *et al.* (1991) [9] also reported lower AUC values following administration of danofloxacin at the same dose level. In contrast, a comparable AUC of $15.6 \mu\text{g}\cdot\text{h/mL}$ was reported by Lopez *et al.* (2015) [11] following intramuscular administration of danofloxacin at a similar dose rate of 5 mg/kg body weight in horses. In addition to the administered dose, AUC is influenced by drug absorption, systemic clearance, and excretory efficiency, affecting overall drug exposure (Toutain *et al.*, 2010) [19]. The mean residence time (MRT) was $7.44 \pm 1.03 \text{ h}$ and $9.90 \pm 0.47 \text{ h}$ following intravenous and intramuscular administration, respectively. Lower MRT values have been reported at a reduced dose of 1.25 mg/kg , including 3.20 h (Aliabadi & Lees, 2003) [2] and 1.25 h (IV) and 6.35 h (IM) by Shem-Tov *et al.* (1998) [17]. In the present study, absolute bioavailability (%F) following intramuscular administration ranged from 77.04% to 110.64%, with a mean value of 96.83%. A higher bioavailability (124%) has been reported at an increased dose of 8 mg/kg in premature calves (Corum *et al.*, 2019) [5], whereas %F values of 76.3% and 101% were reported at a lower dose of 1.25 mg/kg in cattle calves (Aliabadi & Lees, 2003; Giles *et al.*, 1991) [2, 9]. Comparable bioavailability values have also been documented in sheep (95.71%) by McKellar *et al.* (1998) [13], horses (100%) by Lopez *et al.* (2015) [11], and pigs (95.2%) by Zhou *et al.* (2022) [23].

Conclusions

The results of the present investigation demonstrate the existence of breed-related differences in the pharmacokinetic behavior of danofloxacin in Kankrej cattle, indicating the need for further studies to evaluate the underlying mechanisms contributing to these variations. These variations in pharmacokinetic parameters should be taken into account when designing an appropriate dosage regimen for the drug. Danofloxacin showed high systemic availability following intramuscular administration. A single administration of 5.0 mg/kg body weight maintained plasma concentrations within the therapeutic range for approximately 24 h after both intravenous and intramuscular

dosing. Nevertheless, comprehensive pharmacokinetic-pharmacodynamic (PK-PD) integration studies are necessary to assess the adequacy of the current dosing regimen for achieving optimal PK-PD targets against susceptible bacterial pathogens with established minimum inhibitory concentration values in cattle calves.

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