Comparison of plasma pharmacokinetics and bioequivalence of ceftiofur sodium in cattle after a single intramuscular or subcutaneous injection

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Brown, S. A., Chester, S. T., Speedy, A. K., Hubbard, V. L., Callahan, J. K., Hamlow, P. J., Hibbard, B., Robb, E. J. Comparison of plasma pharmacokinetics and bioequivalence of ceftiofur sodium in cattle after a single intramuscular or subcutaneous injection. *J. vet. Pharmacol. Therap.* **23,** 273–280.

Ceftiofur sodium, a broad-spectrum cephalosporin, is active against gram-positive and gram-negative pathogens of veterinary importance. This study was designed to compare the bioequivalence of the sodium salt in cattle after a single intramuscular (i.m.) or subcutaneous dose (s.c.) of 2.2 mg ceftiofur equivalents/kg body weight. The criteria used to evaluate bioequivalence were (1) the area under the curve from time of injection to the limit of quantitation (LOQ) of the assay ($AUC_{0\text{-LOQ}}$), and (2) time concentrations remained above 0.2 μg/mL ($t_{>0.2}$).

Twelve crossbred beef cattle were enrolled in a three-period, two-treatment crossover trial, with a minimum 2-week washout period between doses of 2.2 mg ceftiofur equivalents/kg. Blood samples were collected serially for up to 72 h post-injection. Plasma samples were then analyzed using a validated assay that measures ceftiofur, and all desfuroylceftiofur-related metabolites, by high-performance liquid chromatography (HPLC) as the stable derivative, desfuroylceftiofur acetamide.

A maximum plasma concentration (C_{max}) of 13.9 ± 3.55 μg/mL was observed from $0.67 - 2.0$ h after i.m. administration, whereas a C_{max} of 13.6 ± 3.85 μg/mL was observed from 0.67–3.0 h after s.c. administration. The $AUC_{0\text{-LOQ}}$ was 108 ± 35.0 μg·h/mL after i.m. dosing, compared with 105 ± 29.8 μg·h/mL after s.c. dosing. The pre-established criterion for equivalence of the $AUC_{0\text{-LOQ}}$ for the i.m. and s.c. routes of administration was satisfied. The $t_{>0.2}$ was 49.2 ± 8.55 h after i.m. administration, compared with 47.0 ± 9.40 h after s.c. administration. The pre-established criterion for equivalence of the $t_{>0.2}$ for i.m. and s.c. administration was satisfied.

The equivalence of $AUC_{0\text{-LOQ}}$ and $t_{>0.2}$ for i.m. and s.c. administration of 2.2 mg ceftiofur equivalents (CE)/kg doses of ceftiofur sodium suggest similar therapeutic efficacy and systemic safety for the two routes of administration.

(Paper received 6 July 1999; accepted for publication 3 November 1999)

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INTRODUCTION

Ceftiofur sodium, a broad-spectrum cephalosporin, is active against gram-positive and gram-negative pathogens of veterinary importance, including β-lactamase-producing strains, both *in vitro* and *in vivo* (Yancey *et al.*, 1987). Like other cephalosporins, ceftiofur is bactericidal *in vitro*, resulting from inhibition of cell wall synthesis. The drug is approved for treatment of respiratory diseases of cattle, associated with *Pasteurella haemolytica* (*Mannheimia* spp), *P. multocida* and *Haemophilus somnus*, at doses of $1.1 - 2.2$ mg ceftiofur equivalents (CE)/kg body weight, administered intramuscularly (i.m.) for 3–5 days (FDA, 1988, 1991). Although injection site blemishes rarely occur with i.m. administration, escalating concern in the beef cattle industry over potential blemishes associated with i.m. administration of any product make subcutaneous (s.c.) administration an attractive alternative.

Ceftiofur sodium, regardless of its route of administration, is rapidly metabolized to desfuroylceftiofur. The plasma half-life after intravenous dosing in cattle is 7 min (Banting *et al.*, 1989). Parent ceftiofur is undetectable after 1 h in both cattle and swine (Brown *et al.*, 1991). Desfuroylceftiofur is the primary metabolite and the active moiety for both routes of administration.

Efficacy of β-lactam antibiotics is more closely related to time plasma concentrations that remain above a threshold, typically a multiple of the minimum inhibitory concentration (MIC) for the target pathogens, than to maximum plasma or tissue concentrations (Eagle *et al.*, 1953; Joiner *et al.*, 1982; Frimodt-Møller *et al.*, 1986; Peterson *et al.*, 1984; Frimodt-Møller *et al.*, 1987; Kays *et al.*, 1991). For that reason, bioequivalence was defined as the time concentrations of ceftiofur and metabolites (measured as desfuroylceftiofur acetamide by high-performance liquid chromatography (HPLC)) in plasma remained above a threshold concentration (0.2 μg/mL) set several-fold above the MIC.

This study compared the concentration-time profiles of ceftiofur sodium after single i.m. or s.c. injections. The variables assessed for bioequivalence were (1) the area under the concentration-time curve to the limit of quantitation of the assay (AUC_{LOQ}), and (2) time above 0.20 μg/mL ($t_{>0.2}$), with maximum concentration (C_{max}) and time of observed maximum concentration (t_{max}) being recorded, but not used in decision making.

METHODS

Experimental animals and design

Twelve crossbred beef cattle (six steers and six heifers) were enrolled in a three-period, two-treatment crossover trial (ABB/BAA), with a minimum 2-week washout period between doses of 2.2 mg CE/kg as ceftiofur sodium (NAXCEL®/EXCENEL™ Sterile Powder, Pharmacia & Upjohn Company, Kalamazoo, MI, USA). The dose level represents the upper end of the approved dosage range in the US. Treatment A, using the s.c. route of administration, and treatment B used the i.m. route of administration. The i.m. injection into the neck region or s.c. injection into the manually tented skin of the neck region was performed using a 16-gauge, 1 1/2 inch needle. Injection sites alternated between the animals' right and left sides during the three periods. Study protocols were approved by the appropriate institutional animal care and use committees.

Blood sampling and analysis

Blood samples of 14 mL each were collected by venipuncture before drug administration, and at 20 min, 40 min, 1, 1.5, 2, 3, 4, 8, 16, 24, 36, 48, 60 and 72 h after each injection of 2.2 mg CE/kg. Each blood sample was collected in a heparinized container and stored on ice until processed into plasma. Plasma samples were analyzed using a validated assay that measures ceftiofur and all desfuroylceftiofur-related metabolites with an intact β-lactam ring by HPLC (Jaglan *et al.*, 1990; Hamlow, 1995). Because standard solutions were derived from ceftiofur reference standards, assay results were reported as CE in μg/ml, and summarized as means $\pm$ the standard deviation (SD). The limit of quantitation (LOQ) for the assay is 0.15 μg CE/mL of plasma. Values $<$ LOQ were treated as 0 in the summary statistics.

Pharmacokinetic analysis

The trapezoidal rule was used to determine the area under the concentration-time curve from time 0 (the time of injection) to the last observation above the LOQ ($AUC_{0\text{-}LOQ}$), the *AUC* from 0 to infinity ($AUC_{0-\infty}$)using the terminal rate constant (β), and the terminal phase half-life ($t_{1/2-\beta}$) determined from MODEL 200 (trapezoidal method) from PCNONLIN, Version 4.0 (Statistical Consultants, Inc., Lexington, KY), observed maximum plasma concentration (C_{max}) and time of observed maximum plasma concentration (t_{max}). The time plasma concentrations remained above 0.2 μg/mL ($t_{>0.2}$) was determined by linear regression of the terminal portion of the ln (concentration) vs. time curve or use of a predictive pharmacokinetic equation of the following forms:

$$C_p = \sum_{i=1}^{Z} C_i e^{-\lambda_i t}$$

where λ_i, are macrorate constants describing the apparent absorption and the various apparent elimination rate constants; C_i, are coefficients; and t is the time in hours after drug administration. From the macrorate constants, the corresponding half-lives were calculated.

Fitting of those equations was achieved using nonlinear least squares regression analysis, also using PCNONLIN, Version 4.0, with a weighting factor of concentration^{-1}. Goodness of fit between the two models was determined by Akaike's information criteria, residual trend evaluation of the terminal concentration-time points, and precision of parameter estimates.

Statistical analysis

To compare i.m. vs. s.c. administration, the study was designed as a three-period, two-treatment crossover design. The addition of an extra period dramatically reduces the two major drawbacks of the 2×2 crossover design—its lack of power in detecting (first order) carryover and the requirement that no carryover be present for the treatment comparisons to be unbiased. The extended period design is 'balanced' with respect to carryover in that each treatment not only follows the other treatment, but also follows itself. The test for carryover for the extended period design uses within-animal variance while the 2×2 crossover design uses between-animal variance. In addition, the test of treatment differences is independent of carryover, so the test is unbiased, even in its presence. The additional

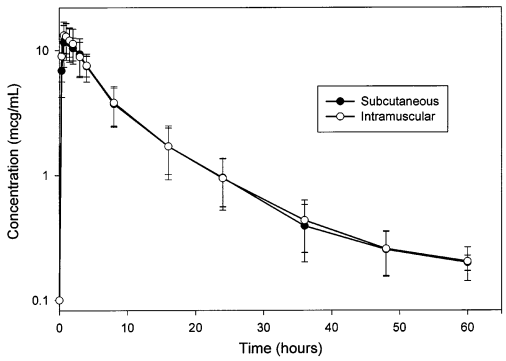

Fig. 1. Mean plasma concentrations (and SDs) of ceftiofur and desfuroylceftiofur-related metabolites over time in cattle following a single i.m. or s.c. injection of 2.2 mg CE/kg body weight of ceftiofur sodium.

observations beyond the 2×2 crossover design produce a 25% reduction in the variance of treatment difference, as well as twice the number of degrees of freedom for error, compared with the 2×2 crossover design (Chow & Liu, 1992). Effects were considered statistically significant at an α level of 0.05 for a given response variable. The power associated with testing our hypothesis using $\alpha = 0.05$ was calculated using both point estimate and upper 75% confidence limit for predetermined equivalence criteria.

Equivalence analysis

Two criteria were used to assess therapeutic equivalence: (1) *AUC* from time of injection to the LOQ of the assay ($AUC_{0\text{-LOQ}}$), and (2) time concentrations remained above 0.2 μg/mL ($t_{>0.2}$). C_{max} and t_{max} were tabulated but not used in the assessment of bioequivalence. The criteria for accepting bioequivalence was if the 90% confidence interval of the difference between the test formulation (s.c. administration) and the reference formulation (i.m. administration) for the variables $AUC_{0\text{-LOQ}}$ and $t_{>0.2}$ resided within $\pm$ 20% of the least square mean of the respective variables for the reference route of administration (i.m.).

RESULTS

All of the animals remained healthy throughout the study and no adverse reactions were observed. Fig. 1 shows the logarithm of plasma concentrations vs. time ($\bar{X} \pm$ SD) for ceftiofur sodium after i.m. and s.c. administration. Individual animal pharmacokinetic data are provided in Tables 1–4. These data are summarized for i.m. and s.c. routes in Table 5. The C_{max} of 13.9 ± 3.55 μg/mL was observed from $0.67 - 2.0$ h after i.m. administration, whereas a C_{max} of 13.6 ± 3.85 μg/mL was observed from $0.67 - 3.0$ h after s.c. administration. For C_{max} the 90% non-parametric confidence interval of the difference between the two routes of administration (-4.50 to 2.98 μg CE/mL) was not contained within the interval of $\pm$ 20% of the i.m. least square mean ($\pm$ 2.79 μg CE/mL). However, the *t*-based 90% confidence interval was -1.33–2.15 μg CE/mL, completely contained within the $\pm$ 20% interval. The discrepancy between the two confidence intervals was a result of two outliers for which there was no apparent cause.

The $AUC_{0\text{-LOQ}}$ by the trapezoidal method was 108 ± 35.0 μg·h/mL after i.m. dosing, compared with 105 ± 29.8 μg·h/mL after s.c. dosing. For AUC_{0-LOQ} the 90% non-parametric confidence interval of the difference between the two routes of administration was -10.1–20.4 μg·h/mL, which was contained within the $\pm$ 20% range of the mean for i.m. administration ($\pm$ 21.9 μg·h/mL). Therefore, the criterion for equivalence of the $AUC_{0\text{-LOQ}}$ for the i.m. and s.c. routes of administration was satisfied.

The observed t_{max} ranged from 0.67–2 h after i.m. administration, whereas the observed t_{max} after s.c. administration ranged from 0.67–3 h. These values were not significantly different between the two routes of administration.

The terminal phase $t_{1/2}$, determined by the terminal data points above the LOQ using the output from the trapezoidal summation method, after i.m. administration was 10.7 ± 3.11 h (harmonic mean = 10.0 h), compared with 9.84 ± 1.97 (harmonic mean = 9.47 h) after s.c. administration.

The plasma concentration vs. time data for both routes of administration were best described by a triexponential model with two disappearance rate constants and a single apparent absorption rate constant. From those data, the terminal elimination half-life was 13.4 ± 4.95 h after s.c. administration, compared with 12.7 ± 3.15 h after i.m. administration. The model-derived *AUC* comparisons were similar to those determined by the trapezoidal method, indicating that the data were well-behaved with regard to pharmacokinetic analysis.

The $t_{>0.2}$ was 49.2 ± 8.55 h after i.m. administration, compared with 47.0 ± 9.40 h after s.c. administration. For $t_{>0.2}$ the 90% non-parametric confidence interval of the difference between the two routes of administration (-2.5 to 10.0 h) was contained within $\pm$ 20% of the least square mean $t_{>0.2}$ after i.m. administration ($\pm$ 10.0 h). Therefore, the criterion for equivalence of the $t_{>0.2}$ for i.m. and s.c. administration was satisfied.

DISCUSSION

Conventional bioequivalence studies evaluate peak concentration (C_{max}) and time to maximum concentration (t_{max}), in addition to $AUC_{0\text{-LOQ}}$, as the decision criteria regarding bioequivalence (Gibaldi & Perrier, 1982; Chow & Liu, 1992). However, the literature for β-lactam antibiotics repeatedly reports that efficacy is not related as closely to maximum plasma or tissue concentrations as to the time concentrations, which remain above a certain threshold. Typically, the threshold is a multiple of the minimum inhibitory concentration (MIC) for the target pathogens (Eagle *et al.*, 1953; Joiner *et al.*, 1982; Peterson *et al.*, 1984; Frimodt-Møller *et al.*, 1986, 1987; Kays *et al.*, 1991). We believe that the time during which the concentration of ceftiofur and metabolites in plasma that is above such a

Table 1. Pharmacokinetic values for ceftiofur and desfuroylceftiofur-related metabolites obtained by the trapezoidal method, after s.c. administration of ceftiofur sodium in cattle as a single dose of 2.2 mg CFAE/kg

Animal no.	Period	t_{max} (h)	C_{max} (μg/mL)	λ_z (h^{-1})	$t_{1/2}$ (h)	AUC_{0-LOQ} (μg · h/mL)	$AUC_{0-\infty}$ (μg · h/mL)	$AUMC_{0-LOQ}$ (μg · h^2/mL)	$AUMC_{0-\infty}$ (μg · h^2/mL)	MRT_{0-LOQ} (h)	$MRT_{0-\infty}$ (h)
584	1	0.67	23.9	0.0685	10.1	128	131	1000	1210	7.88	9.25
587		1	11.5	0.0875	7.92	74.1	76.7	544	666	7.34	8.68
590		1.5	12.5	0.0528	13.1	107	112	986	1300	9.23	11.7
591		1	13.2	0.0647	10.7	123	126	1520	1750	12.4	13.9
592		1	13.6	0.0874	7.93	78.5	81.6	635	782	8.08	9.58
593		0.67	8.05	0.0817	8.49	64.5	66.5	651	770	10.1	11.6
582	2	1	18.3	0.0593	11.7	158	162	1930	2240	12.2	13.8
586		1	12.4	0.109	6.35	77.6	79.5	516	603	6.64	7.58
589		2	10.5	0.0749	9.26	75.2	79.6	599	818	7.97	10.3
594		1	10.6	0.0685	10.1	106	108	1020	1180	9.69	10.9
595		0.67	16.4	0.0787	8.81	107	109	1040	1160	9.71	10.7
596		1.5	13.5	0.0503	13.8	149	154	1870	2260	12.5	14.7
582	3	1	15.3	0.0584	11.9	156	159	1680	1930	10.8	12.1
586		1	12.2	0.0708	9.79	86.5	89.5	590	740	6.82	8.26
589		2	9.10	0.0836	8.29	89.7	92.8	728	877	8.11	9.45
594		1.5	11.2	0.0875	7.92	95.3	97.1	831	936	8.72	9.64
595		0.67	13.2	0.0720	9.63	79.2	81.8	739	899	9.33	11.0
596		3	18.9	0.0610	11.4	129	132	1430	1660	11.1	12.6
Mean		N/A†	13.6	0.0732	9.84	105	108	1020	1210	9.36	10.9
SD		N/A	3.85	0.0148	1.97	29.8	30.1	469	536	1.86	2.01

† N/A = Not applicable. Median value for t_{max} = 1 h.

Table 2. Pharmacokinetic values for ceftiofur and desfuroylceftiofur-related metabolites obtained by the trapezoidal method after i.m. administration of ceftiofur sodium in cattle as a single dose of 2.2 mg CFAE/kg

Animal no.	Period	t_{max} (h)	C_{max} (μg/mL)	λ_z (h^{-1})	$t_{1/2}$ (h)	AUC_{0-LOQ} (μg · h/mL)	$AUC_{0-\infty}$ (μg · h/mL)	$AUMC_{0-LOQ}$ (μg · h^2/mL)	$AUMC_{0-\infty}$ (μg · h^2/mL)	MRT_{0-LOQ} (h)	$MRT_{0-\infty}$ (h)
582	1	0.67	18.4	0.0546	12.7	164	168	2200	2530	13.4	15.1
586		0.67	11.1	0.0855	8.11	62.4	64.2	513	618	8.22	9.64
589		0.67	10.6	0.0632	11.0	66.3	70.4	503	717	7.60	10.2
594		0.67	13.1	0.0756	9.16	93.5	96.0	914	1070	9.77	11.1
595		0.67	16.5	0.0728	9.53	117	120	1270	1450	10.8	12.1
596		0.67	17.2	0.0620	11.2	147	151	1670	1930	11.3	12.8
584	2	0.67	14.4	0.104	6.64	69.8	72.0	504	604	7.22	8.40
587		1	12.1	0.0862	8.04	85.2	88.6	656	819	7.70	9.24
590		1	15.8	0.0624	11.1	119	123	1040	1270	8.76	10.3
591		0.67	18.6	0.0696	9.96	165	168	1820	2000	11.0	12.0
592		2	11.0	0.0339	20.5	87.8	93.3	709	1130	8.07	12.1
593		0.67	7.72	0.0526	13.2	96.0	99.5	1280	1560	13.4	15.7
584	3	1	11.0	0.0584	11.9	96.6	100	912	1130	9.43	11.3
587		1	15.0	0.0514	13.5	124	127	1220	1460	9.89	11.5
590		0.67	16.7	0.0881	7.87	110	112	839	950	7.62	8.48
591		2	19.7	0.0831	8.34	176	179	1600	1820	9.13	10.2
592		0.67	12.6	0.0729	9.51	78.4	81.0	661	818	8.43	10.1
593		2	8.56	0.0655	10.6	93.1	96.4	982	1190	10.6	12.4
Mean		N/A†	13.9	0.0690	10.7	108	112	1070	1280	9.57	11.3
SD		N/A	3.55	0.0167	3.11	35.0	35.1	492	531	1.87	1.98

† N/A = Not applicable. Median value for $t_{max} = 0.67$ h.

Table 3. Pharmacokinetic values for ceftiofur and desfuroylceftiofur-related metabolites obtained by the predictive equation after s.c. administration of ceftiofur sodium in cattle as a single dose of 2.2 mg CFAE/kg

Animal no.	Period	C_1 (μg/mL)	C_2 (μg/mL)	k_a (h^{-1})	λ_1 (h^{-1})	λ_2 (h^{-1})	*AUC* (μg · h/mL)	*AUMC* (μg · h^2/mL)	MRT (h)	$t_{1/2-k_a}$ (h)	$t_{1/2-1}$ (h)	$t_{1/2-2}$ (h)	$t_{>0.2}$ (h)
584	1	15.0	3.74	4.86	0.254	0.0612	116	1230	10.6	0.143	2.73	11.3	47.9
587		13.9	3.34	1.88	0.362	0.0756	73.5	686	9.33	0.368	1.91	9.17	37.2
590		15.0	2.63	1.68	0.231	0.0509	106	1290	12.2	0.412	3.00	13.6	50.6
591		9.94	1.09	2.46	0.108	0.0299	124	2060	16.7	0.282	6.42	23.2	56.5
592		15.0	3.40	1.77	0.404	0.0685	76.4	811	10.6	0.391	1.72	10.1	41.4
593		6.43	0.450	4.79	0.125	0.0250	68.0	1130	16.6	0.145	5.55	27.7	32.1
582	2	15.0	3.67	2.49	0.176	0.0473	155	2120	13.6	0.278	3.95	14.6	61.4
586		15.0	2.00	2.83	0.299	0.0620	76.5	687	8.98	0.245	2.32	11.2	37.2
589		12.4	1.51	1.52	0.241	0.0418	78.4	1070	13.7	0.454	2.88	16.6	48.4
594		10.8	5.28	1.82	0.276	0.0712	104	1180	11.3	0.381	2.51	9.73	46.0
595		15.0	2.80	6.03	0.408	0.0596	80.8	879	10.9	0.115	1.70	11.6	44.3
596		10.7	3.65	3.10	0.148	0.0444	150	2340	15.6	0.224	4.69	15.6	65.5
582	3	15.0	4.24	2.06	0.180	0.0536	153	1930	12.6	0.336	3.85	12.9	57.0
586		15.0	3.03	1.79	0.281	0.0748	83.9	726	8.66	0.386	2.47	9.26	36.3
589		15.0	4.16	0.870	0.263	0.0770	89.1	895	10.0	0.797	2.64	9.00	39.4
594		15.0	3.14	1.35	0.259	0.0648	92.8	960	10.3	0.514	2.67	10.7	42.5
595		13.5	2.72	7.61	0.373	0.0567	82.2	945	11.5	0.0910	1.86	12.2	46.1
596		13.8	3.72	1.44	0.225	0.0512	122	1680	13.8	0.480	3.08	13.5	57.1
Mean	N/A[†]	13.4	3.03	2.80	0.256	0.0564	102	1260	12.0	0.336	3.11	13.4	47.0
SD	N/A	2.45	1.19	1.83	0.0900	0.0150	28.8	535	2.48	0.172	1.32	4.95	9.40

[†] N/A = Not applicable. Median value for $t_{max} = 1$ h.

Table 4. Pharmacokinetic values for ceftiofur and desfuroylceftiofur-related metabolites obtained by the predictive equation after i.m. administration of ceftiofur sodium in cattle as a single dose of 2.2 mg CFAE/kg

Animal no.	Period	C_1 (μg/mL)	C_2 (μg/mL)	k_a (h^{-1})	λ_1 (h^{-1})	λ_1 (h^{-1})	*AUC* (μg · h/mL)	*AUMC* (μg · h^2/mL)	MRT (h)	$t_{1/2-k_a}$ (h)	$t_{1/2-1}$ (h)	$t_{1/2-2}$ (h)	$t_{>0.2}$ (h)
582	1	15.0	4.05	2.29	0.199	0.0421	163	2660	16.3	0.303	3.48	16.4	71.4
586		11.0	1.28	4.63	0.288	0.0477	62.4	697	11.2	0.150	2.40	14.5	39.0
589		11.2	2.26	3.93	0.336	0.0600	67.5	726	10.8	0.177	2.06	11.6	40.4
594		15.0	4.86	2.39	0.494	0.0688	92.7	1080	11.7	0.290	1.40	10.1	46.4
595		15.0	3.75	5.18	0.321	0.0528	114	1490	13.1	0.134	2.16	13.1	55.6
596		15.0	5.95	2.95	0.296	0.0587	145	1900	13.1	0.235	2.34	11.8	57.8
584	2	12.5	2.99	4.36	0.396	0.0734	68.7	634	9.24	0.159	1.75	9.45	36.9
587		12.1	3.72	2.96	0.311	0.0715	85.5	851	9.95	0.234	2.23	9.69	40.9
590		15.0	3.02	3.34	0.224	0.0552	116	1290	11.1	0.207	3.09	12.6	49.2
591		15.0	7.11	5.12	0.295	0.0621	161	2020	12.5	0.135	2.35	11.2	57.5
592		15.0	0.820	1.87	0.223	0.0304	85.9	1190	13.8	0.371	3.11	22.8	46.4
593		8.74	4.27	1.36	0.337	0.0543	95.1	1520	16.0	0.508	2.06	12.8	56.4
584	3	13.3	3.29	1.79	0.268	0.0589	96.2	1130	11.7	0.388	2.58	11.8	47.5
587		15.0	2.79	2.24	0.205	0.0501	121	1460	12.1	0.310	3.38	13.8	52.6
590		15.0	2.88	3.96	0.231	0.0635	106	993	9.39	0.175	3.00	10.9	42.0
591		15.0	7.50	2.76	0.215	0.0691	170	1890	11.1	0.251	3.22	10.0	52.5
592		13.8	1.78	5.08	0.302	0.0486	79.1	903	11.4	0.136	2.30	14.2	45.0
593		11.5	4.14	1.02	0.272	0.0630	92.8	1190	12.8	0.679	2.55	11.0	48.1
Mean	N/A†	13.6	3.69	3.18	0.290	0.0572	107	1310	12.1	0.269	2.53	12.7	49.2
SD	N/A	1.94	1.81	1.33	0.0739	0.0110	33.6	535	1.93	0.145	0.577	3.15	8.55

† N/A = Not applicable. Median value for $t_{max} = 0.67$ h.

Table 5. Pharmacokinetic values derived by trapezoidal summation for i.m. and s.c. administration of ceftiofur sodium as a single dose of 2.2 mg CE/kg

Pharmacokinetic measure	i.m. administration (mean ± SD)	s.c. administration (mean ± SD)
C_{max} (μg/mL)	13.9 ± 3.55 (13.94†)	13.6 ± 3.85 (13.53†)
$AUC_{0–LOQ}$ (μg · h/mL)	108 ± 35.0 (109.5†)	105 ± 29.8 (103.5†)
$AUC_{0–\infty}$ (μg · h/mL)*	112 ± 35.1	108 ± 30.1
$AUMC_{0–LOQ}$ (μg · h²/mL)	1070 ± 492	1020 ± 469
$AUMC_{0–\infty}$ (μg · h²/mL)*	1280 ± 531	1210 ± 536
t_{max} (h)	0.67 − 2 (range)	0.67 − 3 (range)
$t_{>0.2}$ (h)*	49.2 ± 8.55 (49.9†)	47.0 ± 9.40 (46.4†)
$t_{1/2}$ (h)	10.7 ± 3.11 (harmonic mean = 10.0 h)	9.84 ± 1.97 (harmonic mean = 9.47 h)
$MRT_{0–LOQ}$ (h)	9.57 ± 1.87	9.36 ± 1.86
$MRT_{0–\infty}$ (h)	11.3 ± 1.98	10.9 ± 2.01

* Using a predictive pharmacokinetic equation. † Least square mean.

threshold represents a more appropriate variable with which to assess bioequivalence.

The selected threshold of 0.2 μg/mL exceeded by at least 300% the MIC of ≤ 0.06 μg/mL for ceftiofur against *P. haemolytica* (*Mannheimia spp*), *P. multocida*, and *Haemophilus somnus* (Yancey *et al*., 1987). The MIC for ceftiofur against these, including β-lactamase-producing organisms, is lower than for ampicillin (Yancey *et al*., 1987). Thus, the threshold of 0.2 μg/mL established in these studies afforded a conservative measure of clinical efficacy against these major pathogens in cattle. In addition, the value of 0.2 μg/mL is a value above the limit of quantitation of the HPLC assay (which is 0.15 − 0.18 μg/mL), and, therefore, is a reliable concentration when measured.

As the $t_{>0.2}$ was greater than 47.0 h for ceftiofur sodium given by either route, it is clear that once-daily administration keeps plasma concentrations above the MIC for these targeted cattle pathogens for the recommended dosing interval of 24 h.

CONCLUSIONS

Both decision criteria for acceptance of equivalence of ceftiofur sodium administered i.m. or s.c. were satisfied ($AUC_{0\text{-}LOQ}$ and $t_{>0.2}$). The observed C_{max} was equivalent only when assessed using the *t*-based 90% confidence interval, but not the non-parametric confidence interval. There was insufficient evidence to conclude that t_{max} was equivalent for the two routes of administration. However, C_{max} or t_{max} were not among the decision criteria used. Therefore, s.c. administration is considered to be equivalent to i.m. administration of similar doses of ceftiofur sodium in cattle. This conclusion leads to the interpretation that the two routes of administration would be interchangeable in terms of efficacy, systemic target animal safety, and human food safety (with the possible exception of the injection site) for ceftiofur sodium in cattle.

ACKNOWLEDGMENTS

The authors wish to thank C.R. Dudasko-Timm, Jenny Hartman, and Sarah Staples, MA, ELS, for manuscript preparation.

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