Disposition of Sulfadimethoxine in Cattle: Inclusion of Protein Binding Factors in a Pharmacokinetic Model

D. W. A. BOURNE *§, M. BIALER *¶, L. W. DITTERT *‖,
M. HAYASHI **, G. RUDAWSKY ‡, G. D. KORITZ ‡, and
R. F. BEVILL ‡x

Abstract □ Sulfadimethoxine was administered intravenously and orally to four cattle, and plasma and urine samples were collected at various times postdose. Modeling these data with a linear pharmacokinetic model gave unsatisfactory fits, and the data were subsequently fitted to a one-compartment model with saturable protein binding. The saturable protein binding model included the usual linear excretion and elimination processes as well as protein binding parameters. The values obtained *in vivo* for the binding constant, $5.01 \times 10^4 \, M^{-1}$, and the total protein concentration, $7.89 \times 10^{-4} \, M$, compared favorably with previously reported *in vitro* values. These results indicate that protein binding can be successfully included in a pharmacokinetic model.

Keyphrases □ Sulfadimethoxine—protein binding, pharmacokinetics, cattle □ Protein binding—sulfadimethoxine, pharmacokinetics, cattle □ Pharmacokinetics—sulfadimethoxine, protein binding, cattle

Sulfadimethoxine is a long-acting sulfonamide with bacteriostatic activity against a variety of organisms, and commercial preparations are available for use in chickens, turkeys, dogs, cats, horses, cattle, and humans. Because of the widespread use of sulfonamides, a problem of drug residues in edible food tissues has developed.

To understand the kinetics of sulfonamides in food-producing animals, the disposition of various sulfonamides was investigated in a number of species (1–4). The present study involves the disposition of sulfadimethoxine in cattle after intravenous and oral administration. Previous studies (5–7) investigated sulfadimethoxine kinetics in cattle; however, none of these reports included excretion of metabolites into urine or the extensive binding of sulfadimethoxine to plasma proteins.

EXPERIMENTAL

Animals—Four crossbred heifers[1], 10–12 months old and 161–202 kg, were used. The four animals were weighed and placed in slot-floored metabolism units 3 days prior to drug administration. They received a grain–concentrate mixture in limited amounts and grass, hay, and water *ad libitum* during the acclimatization and experimental periods; however, feed was withheld 24 hr prior to oral dosing. To facilitate urine collection, Foley retention catheters[2] were placed in the urinary bladder of each animal 24 hr prior to drug administration.

Drugs—Sulfadimethoxine[3] was prepared as a 12.5% solution in sterile distilled water. Complete solution of sulfadimethoxine was aided with a sufficient quantity of sodium hydroxide.

Dosing Procedure—Sulfadimethoxine (107 mg/kg of body weight) was administered by rapid intravenous drip into the left jugular vein of each animal. After a washout period of ~3 months, sulfadimethoxine (107 mg/kg of body weight) was administered to each animal *via* stomach tube.

Sample Collection—Blood and urine samples were collected at 0, 0.5,

1, 2, 3, 4, 6, 8, 10, 12, 16, 24, 36, 48, 60, 72, 84, 96, 108, and 120 hr following drug administration. Blood samples were taken from the right jugular vein in heparinized syringes and centrifuged at 3000 rpm for 7 min. The plasma was collected and stored at 4° until assay; all plasma samples were assayed within 1 week. The total volume of urine excreted during each sampling period was recorded before aliquots were taken. Urine samples were stored at −20° until assay.

Assay Methods—The plasma sulfadimethoxine concentration was determined spectrophotometrically following color formation with Bratton–Marshall reagents (2, 8). The concentrations of sulfadimethoxine and metabolites in urine were determined spectrophotometrically following separation by TLC and color formation, as previously described (2).

RESULTS AND DISCUSSION

Analytical Data following Intravenous Administration—The average plasma concentration *versus* time data obtained following intravenous sulfadimethoxine administration to cattle (Table I and Fig. 1) appeared to fall biexponentially with a terminal rate constant of ~0.065 hr^{-1} (biological half-life of 10.7 hr). The area under the average plasma concentration *versus* time curve from 0 to 120 hr was 3030 mg hr/liter.

The average excretion rate of unchanged sulfadimethoxine into urine *versus* time data (Fig. 1) also appeared to fall biexponentially but with
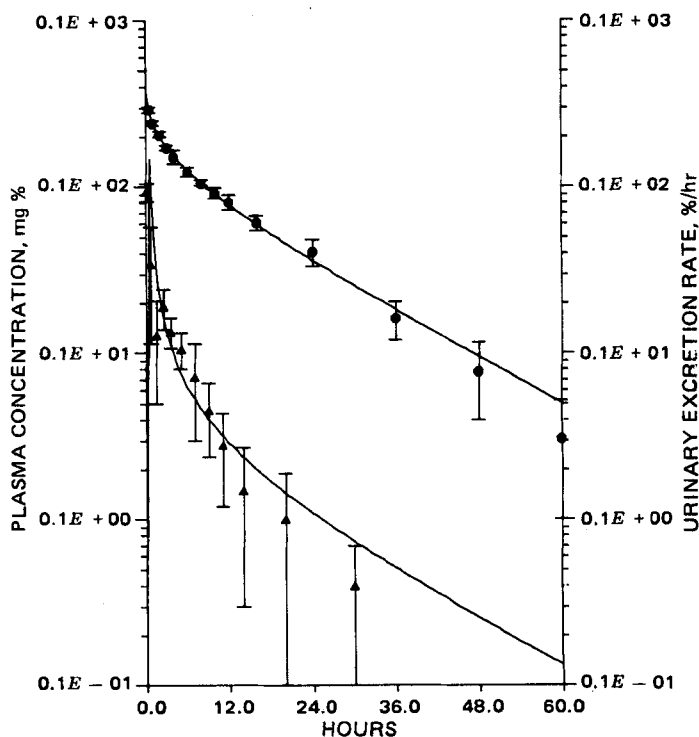

Figure 1—*Semilog plot of plasma concentration (●) versus time and rate of excretion of unchanged sulfadimethoxine in urine (▲) versus time following intravenous sulfadimethoxine administration. Points are the experimentally observed averages (SD as error bars) for four cattle, and lines were calculated.*

Table I—Average Plasma Concentration of Sulfadimethoxine and Cumulative Amount of Sulfadimethoxine, Acetylsulfadimethoxine, and Polar Metabolite Excreted in Urine following Intravenous Sulfadimethoxine Administration

Hours	Plasma Concentration, mg/100 ml	Cumulative Amount Excreted in Urine, % of dose		
		Sulfa-dimethoxine	Acetylsulfa-dimethoxine	Polar Metabolite
0.5	29.3 ± 1.2[a]	4.7 ± 0.6	1.8 ± 0.4	0.2 ± 0.1
1.0	24.3 ± 0.9	6.4 ± 1.6	4.5 ± 0.5	0.3 ± 0.1
2.0	20.8 ± 0.8	7.7 ± 2.3	10.4 ± 1.3	0.6 ± 0.3
3.0	17.3 ± 0.6	9.6 ± 2.6	16.9 ± 1.6	0.9 ± 0.4
4.0	15.3 ± 1.4	11.0 ± 2.5	22.2 ± 2.1	1.2 ± 0.5
6.0	12.4 ± 0.8	13.1 ± 2.5	30.2 ± 1.7	1.9 ± 0.6
8.0	10.6 ± 0.4	14.5 ± 3.1	34.8 ± 2.8	2.3 ± 0.6
10.0	9.2 ± 0.6	15.4 ± 3.3	38.7 ± 3.4	2.4 ± 0.6
12.0	8.2 ± 0.8	16.0 ± 3.6	41.6 ± 3.4	2.6 ± 0.7
16.0	6.1 ± 0.7	16.6 ± 3.9	45.3 ± 4.5	2.8 ± 0.8
24.0	4.1 ± 0.7	17.4 ± 4.1	49.8 ± 5.3	3.0 ± 1.0
36.0	1.6 ± 0.4	17.9 ± 4.4	52.8 ± 6.3	3.4 ± 1.4
48.0	0.8 ± 0.4	17.9 ± 4.4	53.8 ± 7.0	3.5 ± 1.7
60.0	0.3 ± 0.2	—[b]	54.4 ± 7.4	3.7 ± 2.0
72.0	—[c]	—[b]	—[d]	3.8 ± 2.2
84.0	—[c]	—[b]	—[d]	4.0 ± 2.4
96.0	—[c]	—[b]	—[d]	—[e]
108.0	—[c]	—[b]	—[d]	—[e]
120.0	—[c]	—[b]	—[d]	—[e]

[a] One standard deviation (n = 4). [b] No further sulfadimethoxine was detected in urine after 48 hr. [c] Less than 0.1 mg/100 ml. [d] No further acetylsulfadimethoxine was detected in urine after 60 hr. [e] No further polar metabolite was detected in urine after 84 hr.

a slope quite different from that of the plasma data, especially at early time points. The total cumulative amounts of unchanged sulfadimethoxine, acetylsulfadimethoxine, and a polar metabolite excreted into urine were 17.9, 54.4, and 4.0% of the administered dose, respectively (Table I and Fig. 2).

Similar results were obtained for all four individual animals. Results for one animal are shown in Fig. 3.

Analytical Data following Oral Administration—The average plasma concentration *versus* time data obtained following oral sulfadimethoxine administration to cattle are shown in Table II and Fig. 4. The peak plasma concentration was 114 mg/liter at 10 hr, and the area under the plasma concentration–time curve from 0 to 120 hr was 4458 mg hr/liter. The total cumulative amounts of unchanged sulfadimethoxine, acetylsulfadimethoxine, and a polar metabolite excreted into urine were 6.3, 35.6, and 2.1% of the administered dose, respectively (Table II).

Selection of Pharmacokinetic Model—On the basis of plasma concentration data alone, a three-compartment linear model was used
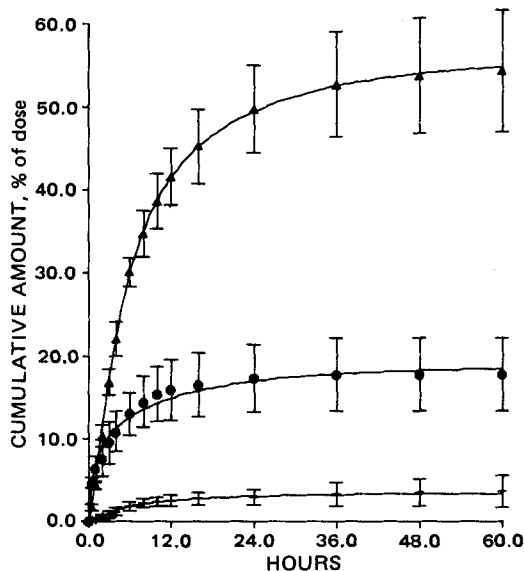

Figure 2—*Plot of cumulative amounts of sulfadimethoxine (●), acetylsulfadimethoxine (▲), and polar metabolite (+) excreted into urine versus time. Points are the experimentally observed averages for four cattle, and lines were calculated.*
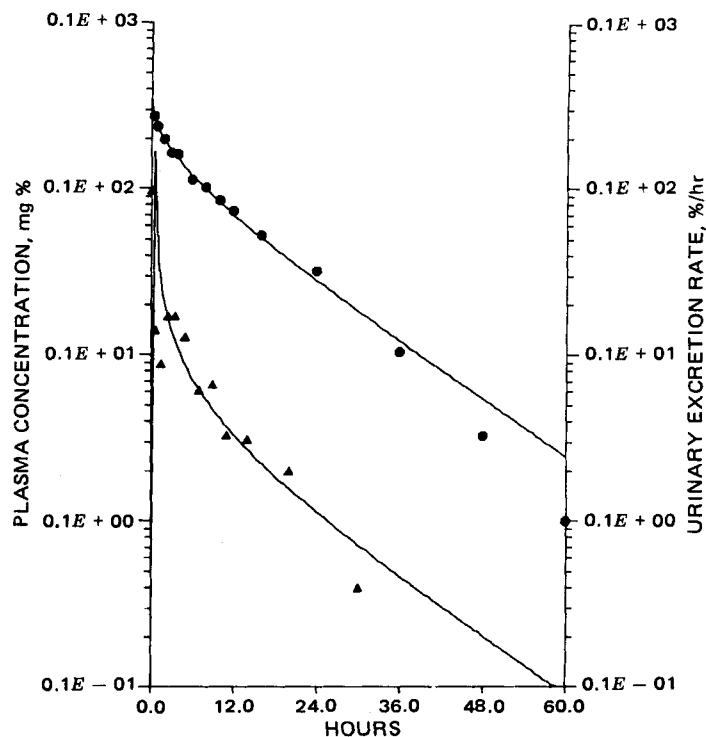

Figure 3—*Semilog plot of plasma concentration (●) versus time and rate of excretion of unchanged sulfadimethoxine in urine (▲) versus time following intravenous administration to one animal. Points were experimentally observed, and lines were calculated.*

to describe the elimination kinetics of sulfadimethoxine in cattle (7). However, a linear model, including the three-compartment model described previously, would predict that the plasma data and the urinary rate of excretion data would appear parallel when plotted semilogarithmically. If linear first-order kinetics apply, the rate of excretion at any time is directly proportional to the plasma concentration. The results obtained (Fig. 1) suggest that there is no constant proportionality between plasma concentration and the rate of excretion data at each time point measured so a simple linear model may not be sufficient to explain the data.

Table II—Average Plasma Concentration of Sulfadimethoxine and Cumulative Amount of Sulfadimethoxine, Acetylsulfadimethoxine, and Polar Metabolite Excreted in Urine following Oral Sulfadimethoxine Administration

Hours	Plasma Concentration, mg/100 ml	Cumulative Amount Excreted in Urine, % of dose		
		Sulfa-dimethoxine	Acetylsulfa-dimethoxine	Polar Metabolite
0.5	1.8 ± 1.4[a]	0.0	0.0	0.0
1.0	4.0 ± 2.0	0.1 ± 0.1	0.1 ± 0.1	0.0
2.0	7.6 ± 2.0	0.1 ± 0.2	0.5 ± 0.4	0.1 ± 0.0
3.0	9.3 ± 1.7	0.2 ± 0.2	1.1 ± 0.6	0.1 ± 0.0
4.0	10.5 ± 1.5	0.3 ± 0.3	2.1 ± 0.9	0.2 ± 0.1
6.0	11.1 ± 1.0	0.5 ± 0.4	3.9 ± 2.4	0.4 ± 0.2
8.0	11.1 ± 0.7	0.8 ± 0.8	5.6 ± 4.6	0.6 ± 0.1
10.0	11.4 ± 1.0	0.9 ± 0.9	7.2 ± 4.6	0.8 ± 0.1
12.0	10.5 ± 0.9	0.9 ± 0.9	8.2 ± 4.5	0.9 ± 0.2
16.0	9.1 ± 1.1	1.1 ± 1.0	9.8 ± 5.3	0.9 ± 0.2
24.0	8.6 ± 1.0	2.0 ± 1.5	18.2 ± 7.3	0.9 ± 0.2
36.0	6.3 ± 1.4	4.6 ± 1.2	27.7 ± 5.1	1.0 ± 0.5
48.0	4.0 ± 0.9	5.9 ± 1.3	32.7 ± 3.8	1.2 ± 0.7
60.0	2.1 ± 0.6	6.1 ± 1.2	34.5 ± 3.6	1.3 ± 1.0
72.0	1.1 ± 0.5	6.2 ± 1.3	35.1 ± 3.8	1.5 ± 1.3
84.0	0.6 ± 0.6	6.3 ± 1.3	35.4 ± 4.1	1.6 ± 1.6
96.0	0.2 ± 0.1	—[b]	35.6 ± 4.4	1.7 ± 1.8
108.0	—[c]	—[b]	—[d]	1.8 ± 1.9
120.0	—[c]	—[b]	—[d]	1.9 ± 2.1
132.0	—[c]	—[b]	—[d]	2.1 ± 2.6
144.0	—[c]	—[b]	—[d]	2.1 ± 2.6

[a] One standard deviation (n = 4). [b] No further sulfadimethoxine was detected in urine after 84 hr. [c] Less than 0.1 mg/100 ml. [d] No further acetylsulfadimethoxine was detected in urine after 96 hr.
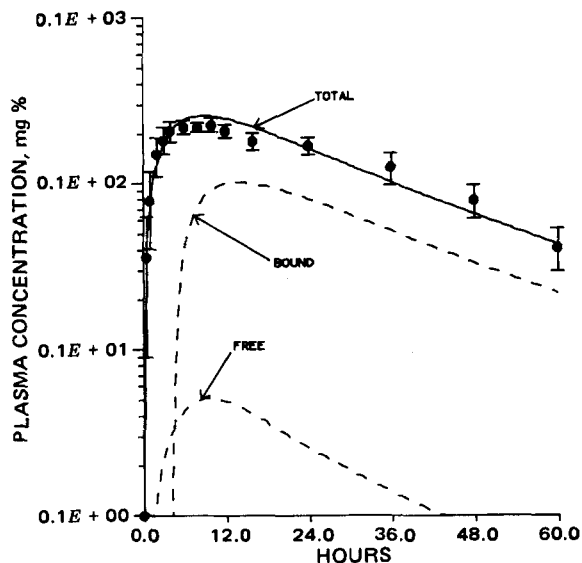
Figure 4—*Semilog plot of plasma concentration (●) versus time following oral sulfadimethoxine administration. Points are the experimentally observed averages for four cattle, and lines were calculated. The points and the solid line were multiplied by 2 to separate these values from the dashed lines representing free and bound drug concentrations.*

Sulfadimethoxine is extensively bound to bovine serum albumin, and the percentage of drug bound to protein decreases as the total concentration of drug in plasma increases (6). Since only free drug can be removed by the kidney or metabolized, protein binding of sulfadimethoxine should extensively affect sulfadimethoxine disposition. Kruger-Thiemer et al. (9) proposed a number of pharmacokinetic models involving protein binding of sulfonamides in humans. By means of computer simulations, these authors fit the plasma concentration *versus* time data to a pharmacokinetic model involving protein binding.

In view of the extensive protein binding of sulfadimethoxine in bovine plasma, it was decided that a model involving protein binding could be used to fit the data. A one-compartment model (Scheme I) was selected as the simplest model involving reversible protein binding.
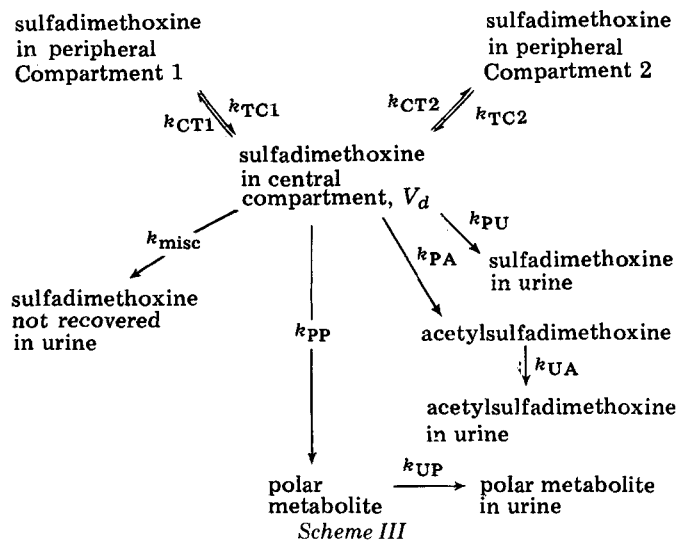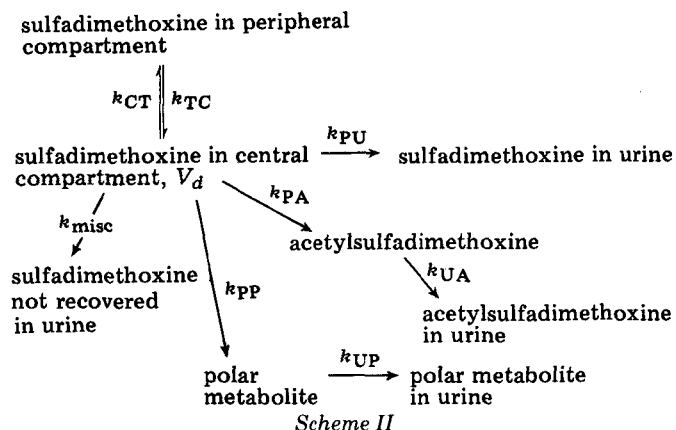

Scheme I
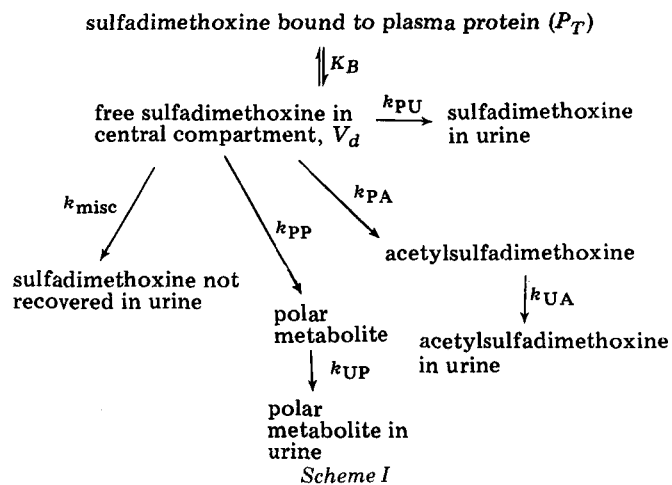
Value K_B is the protein binding constant, P_T is the total concentration of protein binding sites, and V_d is the apparent volume of distribution.

Because drug and metabolite concentrations in urine were measured, this model includes metabolism of sulfadimethoxine and excretion of the two metabolites. All transfer processes in this model are linear, obeying first-order kinetics, with protein binding described by:

$$K_B = [\text{drug bound to protein}]/[\text{free drug}][\text{free protein binding sites}]$$

(Eq. 1)

The data were also fitted to linear two- and three-compartment models

(Schemes II and III, respectively) so that a statistical evaluation of the model shown in Scheme I could be made.

Scheme II

Scheme III

Fitting Data to Pharmacokinetic Model—*Intravenous Study*—The plasma and urine data obtained following intravenous administration of sulfadimethoxine were fitted simultaneously to the model shown in Scheme I and to the linear models shown in Schemes II and III using the digital computer program NONLIN.

An initial estimate of the overall elimination rate constant, k_{el} (= k_{PU} + k_{PA} + k_{PP} + k_{misc}) (Scheme I), was obtained from the slope of the early rate of urinary excretion of unchanged sulfadimethoxine *versus* time data and the estimated concentration of free sulfadimethoxine present at zero time. This value for k_{el} was separated into values for the individual rate constants (k_{PU}, k_{PA}, k_{PP}, and k_{misc}) by means of the total cumulative amounts of sulfadimethoxine excreted as unchanged drug, acetyl metabolite, or polar metabolite into urine data (1).

The free sulfadimethoxine concentration in plasma at different total concentrations was calculated from the data of Silvestri et al. (6) and was used to obtain initial estimates of the binding equilibrium constant, K_B, and the total concentration of protein binding sites, P_T. The initial fitting of the data was carried out with all of the parameters, except k_{UA} and k_{UP}, held fixed at the initial estimate values. Once best fit values of k_{UA} and k_{UP} were obtained, the fitting was continued with progressively more of the parameters allowed to vary, i.e., k_{PU}, k_{PA}, k_{PP}, k_{misc}, K_B, and then P_T. The final best fit values for all parameters of the model in Scheme I are presented in Table III. The close agreement between the model and the experimental data is shown in Figs. 1–3, 5, and 6. The lines were generated using the model shown in Scheme I with the best fit parameter values of Table III.

Initial estimates for k_{el}, k_{CT}, and k_{TC} (Scheme II) and k_{el}, k_{CT1}, k_{TC1}, k_{CT2}, and k_{TC2} (Scheme III) were obtained by "feathering" the plasma concentration–time data. Initial estimates for the other parameters of the linear models were obtained as already described. The fitting process was completed for each model using the average and individual animal data.

The Akaike criterion (11) was used to compare the proposed model,

Table III—Values of Pharmacokinetic Parameters Obtained Using Scheme I following Intravenous Sulfadimethoxine Administration

Parameter	Animal 1	2	3	4	Average	Mean ± SD
k_{PU}, hr^{-1}	1.399	0.382	0.104	0.173	0.443	0.515 ± 0.601
k_{PA}, hr^{-1}	4.300	1.061	0.448	0.322	1.311	1.533 ± 1.873
k_{PP}, hr^{-1}	0.276	0.0452	0.0191	0.0374	0.0824	0.0944 ± 0.122
k_{misc}, hr^{-1}	1.448	0.333	0.152	0.157	0.518	0.523 ± 0.623
k_{el}, hr^{-1}	7.423	1.821	0.723	0.689	2.354	2.664 ± 3.216
K_B, M^{-1}	10.53 × 10⁴	4.66 × 10⁴	2.88 × 10⁴	1.96 × 10⁴	5.12 × 10⁴	5.01 × 10⁴ ± 3.85 × 10⁴
P_T, M	10.65 × 10⁻⁴	8.48 × 10⁻⁴	6.04 × 10⁻⁴	6.40 × 10⁻⁴	8.69 × 10⁻⁴	7.89 × 10⁻⁴ ± 2.13 × 10⁻⁴
k_{UA}, hr^{-1}	0.552	0.433	0.516	0.940	0.412	0.610 ± 0.225
k_{UP}, hr^{-1}	1.023	0.399	1.128	0.232	0.396	0.696 ± 0.446
V_d, liters	49.2	63.8	47.6	53.6	47.4	53.6 ± 7.3
V_d, liter/kg	0.306	0.316	0.317	0.321	0.279	0.315 ± 0.006
AIC^a P_T	141	110	182	76	56	—
AIC 2C	223	198	247	200	219	—
AIC 3C	227	202	252	205	206	—

^{a} Akaike Information Criteria.

Table IV—Values of Pharmacokinetic Parameters Obtained Using Scheme I following Oral Sulfadimethoxine Administration

Parameter	Animal 1	2	3	4	Average	Mean ± SD
k_{PU}, hr^{-1}	0.490	0.129	0.0500	0.0276	0.129	0.17 ± 0.22
k_{PA}, hr^{-1}	2.847	0.993	0.262	0.273	0.970	1.1 ± 1.2
k_{PP}, hr^{-1}	0.0695	0.0648	0.0106	0.0089	0.0409	0.039 ± 0.033
k_{misc}, hr^{-1}	0.825	0.266	0.860	0.0912	0.322	0.32 ± 0.35
k_{el}, hr^{-1}	4.232	1.452	0.409	0.400	1.462	1.62 ± 1.81
k_{UA}, hr^{-1}	6.76	0.251	2.47	0.0944	0.376	2.4 ± 3.1
k_{UP}, hr^{-1}	50.0	50.0	43.7	48.7	40.1	48.1 ± 3.0
Bioavailability	0.553	0.632	0.530	0.650	0.550	0.591 ± 0.059
k_a, hr^{-1}	0.413	0.113	0.155	0.154	0.187	0.209 ± 0.138

incorporating protein binding parameters (Scheme I) with the linear models (Schemes II and III). In each case, the one-compartment saturable protein binding model gave a better fit than either of the two linear models (Table III). The appropriateness of the chosen model is further supported by the reasonable values obtained for K_B and P_T. The average value of K_B obtained in the present study, 5.01 × 10⁴ M^{-1}, compares favorably with values reported by other workers using bovine serum albumin in vitro, 1.5–25 × 10⁴ M^{-1} (12–14). In addition, the average value of P_T, 7.89 × 10⁻⁴ M, compares well with the plasma albumin concentrations previously reported, 4.5–4.7 × 10⁻⁴ M (15, 16).

Oral Study—The plasma and urine data obtained following oral administration of sulfadimethoxine were fitted to the model shown in Scheme I, with an added first-order absorption step. The initial estimate of the absorption rate constant, k_a, was determined by the method of residuals. The bioavailability was calculated as 0.59. When calculated by comparison of the total amount of drug recovered in urine following intravenous and oral administration, ~47% of the dose was absorbed. The ratio of the area under the plasma concentration–time curve could not be used in this study as a measure of bioavailability since the model was nonlinear. The model is consistent with the observed higher area under the curve following the oral dose compared with the intravenous dose. The values of the parameters of Scheme I obtained from the intravenous study were used as initial estimates in fitting the oral study data with the NONLIN computer program. The final best fit values obtained are
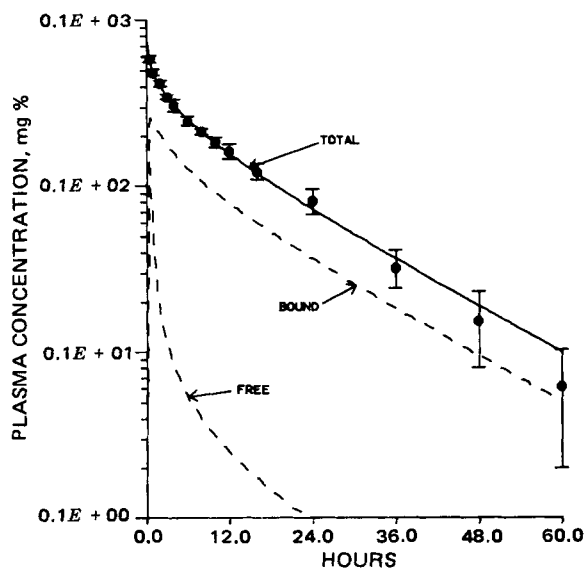

Figure 5—*Semilog plot of plasma concentration (●) versus time following intravenous sulfadimethoxine administration. Points are the experimentally observed averages for four cattle, and lines were calculated. The points and the solid line were multiplied by 2 to separate these values from the dashed lines representing free and bound drug concentrations.*
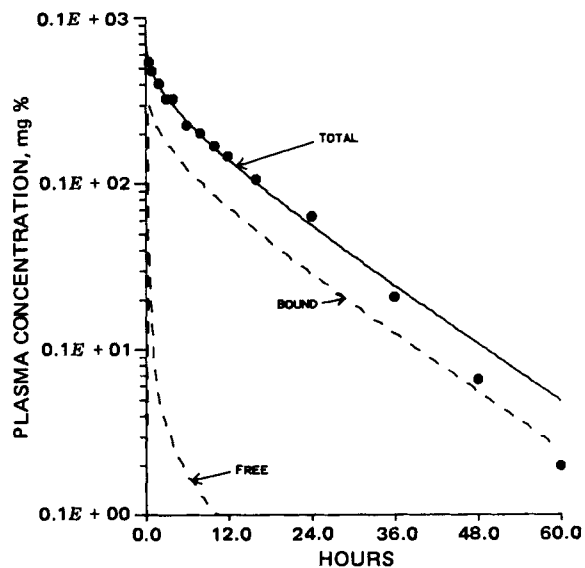

Figure 6—*Semilog plot of plasma concentration (●) versus time following intravenous sulfadimethoxine administration to one animal. Points were experimentally observed, and lines were calculated. The points and the solid line were multiplied by 2 to separate these values from the dashed lines representing free and bound drug concentrations.*
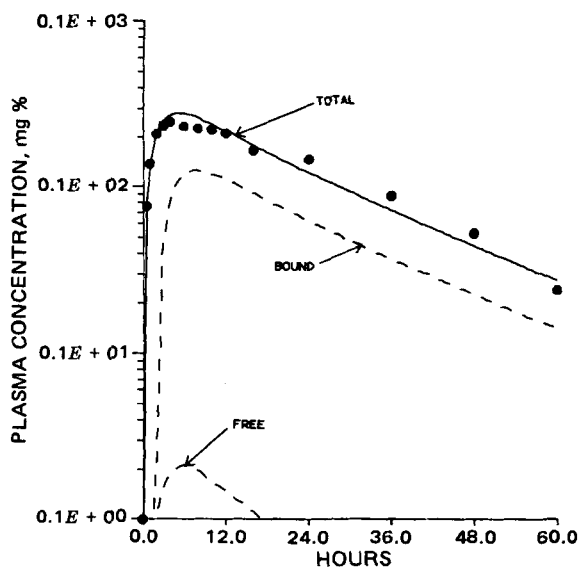
Figure 7—Semilog plot of plasma concentration (●) versus time following oral sulfadimethoxine administration to one animal. Points were experimentally observed, and lines were calculated. The points and the solid line were multiplied by 2 to separate these values from the dashed lines representing free and bound drug concentrations.

presented in Table IV. The lines in Figs. 4 and 7 were calculated using the best fit values of the parameters.

REFERENCES

(1) D. W. A. Bourne, R. F. Bevill, R. M. Sharma, R. P. Gural, and L. W. Dittert, *Am. J. Vet. Res.*, 38, 967 (1977).

(2) R. F. Bevill, R. M. Sharma, S. H. Meachum, S. C. Wozniak, D. W. A. Bourne, and L. W. Dittert, *ibid.*, 38, 973 (1977).

(3) G. D. Koritz, R. F. Bevill, D. W. A. Bourne, and L. W. Dittert, *ibid.*, 39, 481 (1978).

(4) D. W. A. Bourne, L. W. Dittert, G. D. Koritz, and R. F. Bevill, *J. Pharmacokinet. Biopharm.*, 6, 123 (1978).

(5) C. M. Stowe and C. S. Sisodia, *Am. J. Vet. Res.*, 24, 525 (1963).

(6) R. Silvestri, P. Magnifico, and S. Glatstein, *ibid.*, 28, 1783 (1967).

(7) H. G. Boxenbaum and S. A. Kaplan, *J. Pharmacokinet. Biopharm.*, 3, 257 (1975).

(8) J. S. Annino, in "Standard Methods of Clinical Chemistry," vol. 3, Academic, New York, N.Y., 1961, pp. 200–206.

(9) E. Kruger-Thiemer, W. Diller, and P. Bunger, *Antimicrob. Agents Chemother.*, 5, 183 (1965).

(10) C. M. Metzler, G. L. Elfind, and A. J. McEwen, "A User's Manual for NONLIN and Associated Programs, Research Biostatistics," Upjohn Co., Kalamazoo, Mich., 1974.

(11) K. Yamaki, T. Nakagawa, and T. Uno, *J. Pharmacokinet. Biopharm.*, 6, 165 (1978).

(12) P.-L. Hsu, J. K. H. Ma, H. W. Jun, and L. A. Luzzi, *J. Pharm. Sci.*, 63, 27 (1974).

(13) M. Nakagaki, N. Koga, and H. Terada, *Yakugaku Zasshi*, 84, 516 (1964).

(14) I. Moriguchi, S. Wada, and J. Nishizawa, *Chem. Pharm. Bull.*, 16, 601 (1968).

(15) M. S. Spector, "Handbook of Biological Data," Saunders, Philadelphia, Pa., 1956.

(16) E. Genazzani and G. Pagnini, *Am. J. Vet. Res.*, 24, 1212 (1963).

Quantitative Determination of Conjugated Estrogens in Formulations by Capillary GLC

G. K. PILLAI and K. M. McERLANE ˣ

Abstract □ A rapid capillary GLC method for the analysis of conjugated estrogen tablets and injectable formulations is described. The method involves the hydrolytic cleavage of the sodium sulfate ester conjugates by sulfatase enzyme. The free phenolic steroids are reacted sequentially with hydroxylamine hydrochloride and N,O-bis(trimethylsilyl)trifluoroacetamide. The resulting dual derivatives are analyzed on a 15-m glass capillary column wall coated with a cyanopropylmethyl silicone phase.

Keyphrases □ Estrogens, conjugated—quantitative determinations by capillary GLC □ GLC, capillary—quantitative determination of conjugated estrogens □ Steroids—conjugated estrogens, quantitative determination by capillary GLC

Conjugated estrogens is the official name (1) given to a group of closely related steroids derived from the urine of pregnant mares. These naturally occurring, water-soluble salts have been used since 1942 for the treatment of estrogen deficiency symptoms associated with menopause. Only recently, however, have the structures of these steroids been collectively reported (2) and quantitative methods developed (3, 4). For many years, the accepted method of analysis was that contained in USP XVIII (5), which specified colorimetric methods and limits for the major steroids, estrone and equilin, as well as for total estrogen content. An absorbancy ratio requirement was included in the identification tests for an unspecified steroid.

Recent revisions of the USP/NF (1) included a GLC identification test based on a published method (3) but retained the iron–kober chromogenic reagent for the assay of estrone, equilin, and total estrogen content. The identification test requires a prominent peak for one steroid, 17α-dihydroequilin, and this substance probably is the steroid monitored by the previous USP (5) identification test.

BACKGROUND

A consequence of the broad and nonquantitative specifications for the major and minor steroids is that compendial standards may not ensure pharmaceutical equivalence. More rigorous specifications for the indi-